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SAPPORO, JAPAN.

VOL. VI.

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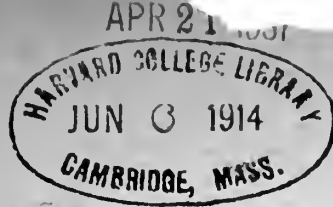
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FURTHER DATA ON THE REDUPLICATION IN SILKWORMS.

By

Y. Tanaka.

1. DISSIMILARITY OF MALE AND FEMALE GAMETIC SERIES IN THE STRIPED-YELLOW COUPLING.

As I have made out in the separate paper now under preparation for press¹, the reduplication in the normal-yellow repulsion and the moricaud-yellow coupling in the silkworm is not of the same intensity in male and female, but it follows quite distinct systems among the opposite sexes. In such cases the reduplication was complete in female, and it was partial in male. The similar sexual dimorphism of gametic series exists also in the striped-yellow coupling which is dealt with in the following pages.

In the previous paper² I have described two families in which coupling occurred between *striped* and *yellow*, the actual numbers being

Lot No.	H. 1'11	Lot No.	S. 5'11
Striped yellow (H. 1'12)	148	Striped yellow	134
Striped white (H. 2'12)	12	Striped white	10
Normal yellow (H. 3'12)	14	Plain yellow	7
Normal white (H. 4'12)	50	Plain white	43
Total	224		194

1. Occurrence of Different Systems of Gametic Reduplication in Male and Female Hybrids. Zeits. f. ind. Abst. u. Vererb. 1914.

2. Gametic Coupling and Repulsion in the Silkworm. This Journal, Vol. V, 1913, pp. 115-148.

Nine matings from H. 1'12 have been reared in 1913, which gave the following results :

	Striped yellow	Striped white	Normal yellow	Normal white	Total
* H. 1-1'12	194	—	80	100	374
H. 1-2'12	86	—	35	—	121
H. 1-3'12	210	—	80	—	290
* H. 1-4'12	192	—	91	81	364
H. 1-5'12	459	—	—	—	459
H. 1-6'12	204	23	12	54	293
H. 1-7'12	155	61	—	—	216
H. 1-8'12	247	26	24	65	362
H. 1-9'12	60	3	10	18	91

Apart awhile from the asterisked families (concerning which see later pages) all of the above results are according to expectation. Totals of those families in which four zygotic forms occurred are

	Striped yellow	Striped white	Normal yellow	Normal white	Total
H. 1-6, 8, 9'12 all together	511	52	46	137	746

Thus existence of a partial coupling between two dominant characters **S** (striped marking) and **Y** (yellow colour) can not be doubted here, and moreover the system of the reduplication seems, at a glance, to be that of 7 : 1 : 1 : 7.¹ But that this was actually not the case has been amply proved by other facts which will be given later.

F₁ striped yellows (H. 29'12) derived from the cross striped yellow ♀ × normal white (*Aojiku*) ♂ gave the following F₂ offspring in the spring 1913.

	Striped yellow	Striped white	Normal yellow	Normal white	Total
H. 29-1'12	240	22	35	63	360

1. Expectation on such basis is 515.7 : 43.7 : 43.7 : 142.9.

H. 29-2'12	140	15	23	48	226
H. 29-3'12	300	35	33	91	459
H. 29-4'12	239	38	40	55	372
H. 29-5'12	342	36	41	94	513
H. 29-6'12	242	50	24	54	370
H. 29-7'12	195	31	29	39	294
H. 29-8'12	275	37	36	63	411
Total	1973	264	261	507	3005

Though the results appear to tempt one to supposition that 4 : 1 : 1 : 4 coupling¹ occurs in this case, this assumption was proved inadequate, as it was the case with the supposition of 7 : 1 coupling in H. 1 families described in the preceding page. The actual feature of the gametic distribution was first revealed by the following data.

a) Results of the cross double recessives (plain or *Kasuri*² whites) ♀ × diheterozygous striped yellows ♂.

	Striped yellow	Striped white	Normal yellow	Normal white	Total
H. 45-2'12 (plain wh. ♀ × H. 29'12 ♂)	88	45	55	97	285
Expect. on 2 : 1 : 1 : 2	95.0	47.5	47.5	95.0	285.0
S. 1'13 (<i>Kasuri</i> wh. ♀ × H. 1-6'12, str. yel. (12'13) ♂) ¹⁷¹		67	67	190	495
Expect. on 3 : 1 : 1 : 3	185.6	61.9	61.9	185.6	495.0
S. 2'13 (ditto)	186	92	101	198	577
Expect. on 2 : 1 : 1 : 2	192.3	96.2	96.2	192.3	577.0
S. 3'13 (ditto)	204	61	80	181	526
Expect. on 3 : 1 : 1 : 3	197.25	65.75	65.75	197.25	526.00
S. 8'13 (<i>Arayahime</i> ♀ × H. 37-1'12, str. yel. (292'13) ♂) ¹¹³		39	33	148	333
Expect. on 3 : 1 : 1 : 3	124.9	41.6	41.6	124.9	333.0

1. Expectation on such assumption will be 1983.30 : 270.45 : 270.45 : 480.80.

2. This is recessive to the normal.

S. 9'13 (ditto)	94	35	39	94	262
Expect. on 3 : 1 : 1 : 3	98.25	32.75	32.75	98.25	262.00
S. 11'13 (<i>Kasuri</i> wh. ♀ × H. 29-8'12, str. yel. (160'13) ♂)	143	73	78	135	429
Expect. on 2 : 1 : 1 : 2	143.0	71.5	71.5	143.0	429.0
Total (S. 1, 3, 8, 9'13)	582	202	219	613	1616
Expect. on 3 : 1 : 1 : 3	606	202	202	606	1616
Total (H. 45-2'12 and S. 2, 11'13)	417	210	234	430	1291
Expect. on 2 : 1 : 1 : 2	430.3	215.2	215.2	430.3	1291.0

From these results we know that the **S-Y** coupling in the male gametic series was neither on 7 : 1 : 1 : 7 nor on 4 : 1 : 1 : 4, but it follows either 3 : 1 : 1 : 3 or 2 : 1 : 1 : 2 system.

b) Results from the cross diheterozygous striped yellows ♀ × double recessives (normal whites) ♂.

	Striped yellow	Normal white	Total
H. 58-1'12 (H. 29'12 ♀ × <i>Chohakuryu</i> ♂)	208	215	423
H. 58-2'12 (ditto)	208	233	441
H. 58-3'12 (ditto)	167	152	319
Total	583	600	1183
Expect. on 1 : 1	591.5	591.5	1183.0

Thus it is evident that a complete repulsion occurs in the female gametic series.

The results above described brought to light the gametic series in **SsYy** animal (into which **S** and **Y** were introduced by the same parent). They do not follow the same system of the reduplication in male and female, but they distinctly differ from each other, namely:

	SY	Sy	sY	sy
Spermatozoa	3	1	1	3
Eggs	2	1	1	2

The union of such gametic series will give rise to the following zygotic series:

Gametic series						Zygotic series			
♀			♂			Str. yel.	Str. wh.	Non-str. yel.	Non-str. wh.
SY	sy	SY	Sy	sY	sy				
1 : 1		3 : 1	1 : 1	1 : 3		11 : 1	1 : 1	1 : 3	
1 : 1		2 : 1	1 : 1	1 : 2		8 : 1	1 : 1	1 : 2	

The ratios 11 : 1 : 1 : 3 and 8 : 1 : 1 : 2 show an approximation to the systems 11.80 : 1 : 1 : 3.27 (=177 : 15 : 15 : 49 which results from the 7 : 1 coupling on both sexes) and 7.33 : 1 : 1 : 1.78 (=66 : 9 : 9 : 16 that is the outcome of the 4 : 1 series on both sexes) respectively. So they seem liable to be mistaken for ordinary 7 : 1 or 4 : 1 coupling. The actual figures are compared with the numbers expected on the above ratios.

	Striped yellow	Striped white	Normal yellow	Normal white	Total
H. 1-6, 8, 9'12 (3 families)	511	52	46	137	746
Expect. on 11 : 1 : 1 : 3 zyg. ser.	512.9	46.6	46.6	139.9	746.0
H. 29-1-8'12 (8 families)	1973	264	261	507	3005
Expect. on 8 : 1 : 1 : 2 zyg. ser.	2003.3	250.4	250.4	500.9	3005.0

The expectation is, as it is shown, fairly realised.

2. TWO NEW INSTANCES OF THE REDUPLICATION IN SILKWORMS.

Besides those cases of gametic reduplication hitherto described by the writer¹ two new examples were found in the silkworm. One was the repulsion between **S** and **Y**, while the other was the coupling between **N** (normal marking) and **Y**.

a) The Repulsion between **S** and **Y**.

This occurred in one (S. 10'13) of the families reared during the summer 1913. The origin of the family in question is as follows.

¹ 1913 l. c. and 1914 l. c.

Table I.

1910	Normal white ♀ × Striped yellow ♂ <i>(Aojiku)</i> (H. 12'10)									
1911	Moricaud yellow ♀ × Normal white ♂ <i>(Aojiku)</i> (H. 14'11)									
1912	Mor. yel. (H. 22'12) × Str. yel. (H. 2'12) 485 (all) 148 12 14 50									
1913, spring	Str. yel. (No. 297'13) × Str. wh. (H. 56'12) 63 59 46 18 13 53									
1913, summer	Str. yel. (S. 10'13) × Kasuri wh. ♀ 82 173 164 69									

Notice:
 Marking.—Str. > Mor. > Nor. > Kas.
 Cocoon colours,—Yel. > Wh.

As above pedigree shows the heterozygous striped yellows (No. 297/13) in the spring 1913¹ have received the stripedness from the mother, and the yellowness from the father. Occurrence of gametic repulsion between these dominant characters may naturally be anticipated² as the result of such crossing, and practically the figures obtained in the succeeding season represent the reduplicated series of the male parent as such, the *Kasuri* white being absolute recessive to the striped yellow.

	Striped yellow	Striped white	Moricaud yellow	Moricaud white	Total
Observed	82	173	164	69	488
Expected on 1 : 2 : 2 : 1 basis	81.3	162.7	162.7	81.3	488.0

Thus it is evident that a partial repulsion of lowest intensity takes place in the *male* gametes of **SsYy**, when the factors **S** and **Y** have been introduced by the different parents. We cannot tell with confidence at present what happens in the *female* heterozygote, though there are some reasons in imagining occurrence of a complete repulsion in it.

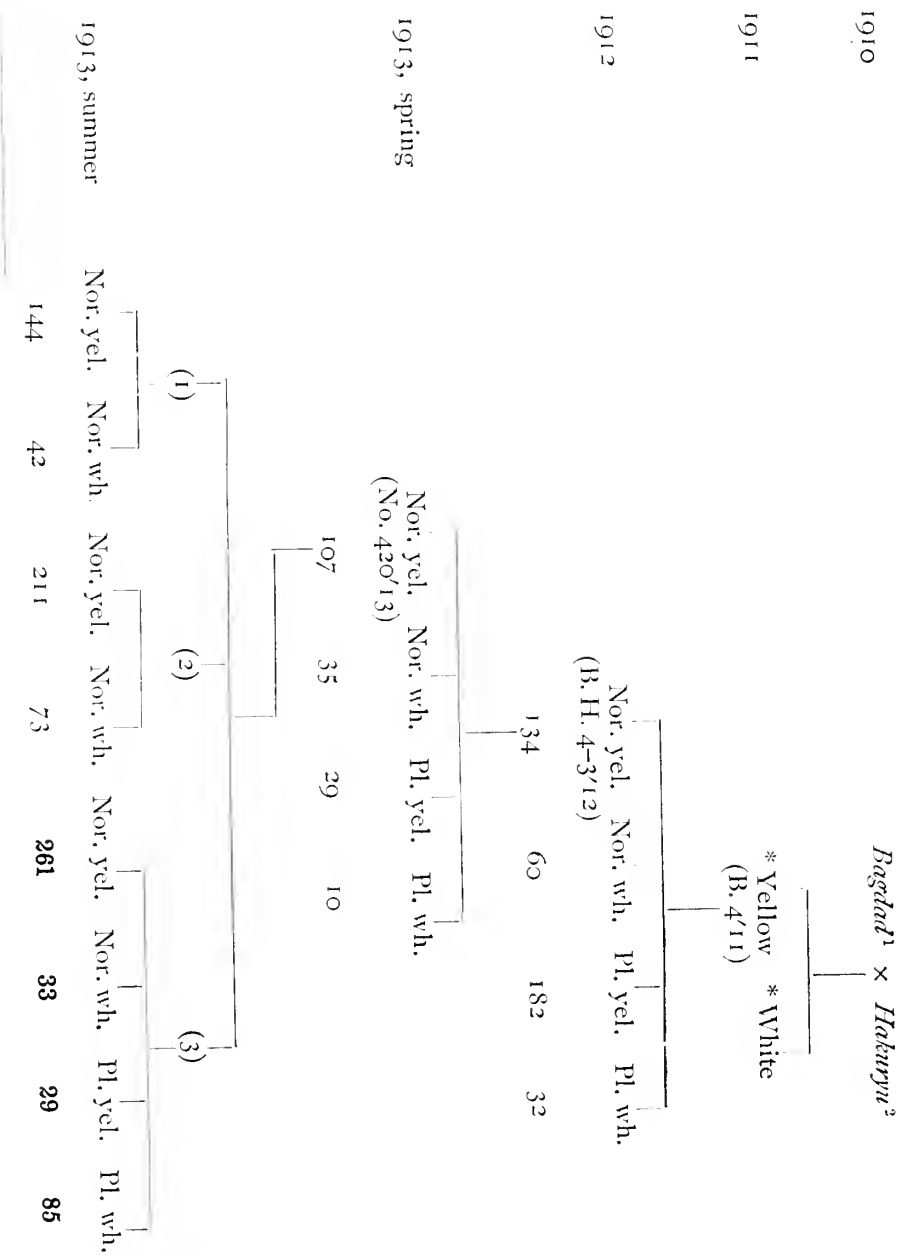
b) The Coupling between **N** and **Y**.

As I have described in the separate paper (1914, l. c.) the repulsion between **N** and **Y** that occurs in the heterozygote ex **NNyy** × **nnYY** is partial in the male, but it is complete in the female. If **N** and **Y** were brought into the 'hybrid by the same parent, on the other hand, what there will be happened? The following result, though insufficient, points to occurrence of at least some couplings, possibly 2 : 1 : 1 : 2 in ♂— and complete in ♀, in such a cross. The pedigree and the actual numbers are diagrammatically given below.

1. The entire result in this generation is easily accounted for as follows. As the gametic series would have been 1 **SY** : 1 **sy** in the mother, and 3 **MY** : 1 **My** : 1 **mY** : 3 **my** in the father (**N**-factor being understood), the offspring should be in the ratio 4 str. yel. : 4 str. wh. : 3 mor. yel. : 1 mor. wh. : 1 nor. yel. : 3 nor. wh. This has been precisely realised, the expectation being 63 : 63 : 47 : 16 : 16 : 47.

2. Couplings arise, as stated elsewhere, in F₂ ex str. yel. × non-str. wh.

Table II.



Y. TANAKA.

1. European dominant white, univoltine.
 2. Japanese normal white, bivoltine.
- * Marking characters as well as the actual numbers were not recorded.

Though the preceding pedigree begins with the cross of dominant and recessive whites, we may put the inhibitor, to which the *Bagdad white* owes its dominancy, out of consideration in the present investigation, because the descendants given above all belong to the yellow series in which the factor in question is necessarily absent.

The result in 1912 is what may be accounted for on the basis that the parents were $nnYy \text{ ♀} \times NnYy \text{ ♂}$, and 1 : 2 : 2 : 1 repulsion occurred in male side. On such assumption we may expect the following zygotic series, which is very close to the actual figures.

	Normal yellow	Normal white	Plain yellow	Plain white	Total
Observed	134	60	182	32	408
Expected	136 (4)	68 (2)	170 (5)	34 (1)	408

The numbers in the spring 1913 precisely accord to 9 : 3 : 3 : 1 series, the expectation being as follows:

	Normal yellow	Normal white	Plain yellow	Plain white	Total
Observed	107	35	29	10	181
Expected	102	34	34	11	181

The result seems suggestive to normal distribution of gametic forms in both sexes. Nevertheless I am inclined, from the following reasons, to believe existence of the underlying reduplication even in this case. In the first place, some of the normal yellows in 1912 (No. 425'13) were possibly produced by union of the gametes Ny and nY , while others were derived from combination of NY and ny ; thus the reduplication in the former was repulsion, and that in the latter was coupling. In the second, the sexual dimorphism of gametic series in the reduplication hitherto described, must be assumed to exist also in the present case. If therefore it chanced a repulsion to occur in one parent, the *father*, and a coupling to take place in another, the *mother*, the gametic series would be

	NY		Ny		nY		ny
♂	1	:	3	:	3	:	1
♀	1			:			1

whence the apparently normal zygotic series, 9 : 3 : 3 : 1, may result.

Lastly we come to the discussion of a family, No. 420-3'13, reared during the summer 1913. The figures in this family seem to tempt one to the supposition that the 4 : 1 : 1 : 4 coupling exists in both parents. Presumably, however, this supposition is not correct, but here the male and female gametic series would have been distinct as it was the case with the striped-yellow and moricaud-yellow couplings. Thus if we assume the gametic series as

	NY		Ny		nY		ny
♂	2	:	1	:	1	:	2
♀	1			:			1

we may expect the following numbers:

	Normal yellow	Normal white	Plain yellow	Plain white	Total
Observed	261	33	29	85	408
Expected	272	34	34	68	408
	(8)	(1)	(1)	(2)	

Conclusive results are to be obtained, however, only from the crosses **NnYy** ♀ × **nnyy** ♂ and the reciprocal, where **NnYy** has been produced by union of **NY** and **ny**.

P. S. In my previous paper (1913, l. c., p. 122 and Table V), I have described a family (II. 17'11) in which apparently complete repulsion occurred between **S** and **Y** in spite of the family has been derived from the cross *Aojiku* (normal white) ♀ × striped yellow ♂. Whether there may exist, in certain occasion, a complete reduplication in *male*, or whether the above result is due to any mistake by which sex-signs have been reversed, is at present uncertain. No similar case has as yet been found in other families.

3. COMPLEX PHENOMENA RELATING TO THE STRIPEDNESS.

Some irregularities observed in the heredity of the striped character are described in the following lines.

a) Occurrence of a Rare Zygotic Series $2 \text{ AB} : 1 \text{ aB} : 1 \text{ ab}$.

In a partial reduplication all possible F_2 zygotic combinations (i. e. four) are produced, while in a complete repulsion three, and in a complete coupling only two phenotypes can occur. Whenever there are only three phenotypes present the zygotic series should be, on an ordinary occasion, $2 \text{ AB} : 1 \text{ Ab} : 1 \text{ aB}$, the last term (ab) being absent.

In my experiments with the silkworm, however, I have met with two families giving an evidence for possibility of $2 \text{ AB} : 1 \text{ aB} : 1 \text{ ab}$ zygotic series when inbred where no Ab individual occurred. An account of these families is given here.

The striped yellows of the family H. 1'12 (Table III), the generation preceding that under consideration, would, as may be inferred from their pedigree, have consisted of four different forms with regards the zygotic constitution, namely SSYY , SSYy , SsYY and SsYy . The offspring of two matings amongst nine matings of these striped yellows showed a peculiar feature as given below¹.

	Striped yellow (SY)	Normal yellow (sY)	Normal white (sy)
H. 1-1'12	194	80	100
H. 1-4'12	192	91	81
Total	386	171	181
Calculated on $2 : 1 : 1$	369.0	184.5	184.5

The Complete pedigree is put forth in the following table.

1. Cf. p. 2.

Striped yellow males (No. 8'13) in one of the families under consideration were crossed with bivoltine *Kasuri* white females, and the offspring were reared during the summer of the same year. The results follow:

	Striped yellow	Striped white	Normal yellow	Normal white	Total
S. 6'13 (<i>Kasuri</i> white ♀ × 8'13 ♂)	162	84	74	178	498
S. 7'13 (ditto)	222	—	188	—	410
S. 12'13 (ditto)	231	—	221	—	452

The matter which became clear to us from these results is nothing but that some of the striped yellow parents were **SsYy**, in which 2 : 1 : 1 : 2 coupling occurred, and some others were **SsYY** in their zygotic constitutions. This gives us, as far as it goes, apparently no clue to solution of the question how such a peculiar system as 2 **AB** : 1 **aB** : 1 **ab** (where **A** stands for **S**, **B** for **Y**) has been brought forth in the preceding generation. Thorough analysis of the case may be expected only after sufficient data have been accumulated.

If we assume a spontaneous elimination of **S**-factor in **SsYy** ♂ the foregoing case may easily be accounted for. But this assumption is evidently premature at present.

b) A Peculiar Type of Gametic Coupling between **S** and **Y**.

As mentioned in the foregoing pages the zygotic series in striped-yellow coupling are, in usual case, as follows:

	Str. yel.		Str. wh.		Nor. yel.		Nor. wh.
	11	:	1	:	1	:	3
or	8	:	1	:	1	:	2

These are effected from the male gametic series 3 : 1 : 1 : 3 and 2 : 1 : 1 : 2 respectively, the female gametes being 1 **SY** : 1 **sy** in both cases. An aberrant phenomenon from this general rule, however, was observed in the families H. 31-2, 3, 4'12 in which only two zygotic forms, double dominant and double recessive, occurred in *equal* numbers.

The actual figures and the origin of the families are given below.

Normal white ♀ × Striped yellow ♂
(*Aojiku*) (H. 1'10)

1910	Normal white ♀ × Striped yellow ♂ (<i>Aojiku</i>) (H. 1'10)																													
1911	Str. yel. (H. 1'11) 161										Nor. yel. 115																			
1912	Str. yel. (H. 1'12) 148			Str. wh. 12			Nor. yel. 14			Nor. wh. 50																				
	(1)			(2)		(3)		(4)			(5)		(6)		(7)		(8)		(9)											
1913, spring	Str. yel.	Nor. yel.	Nor. wh.	Str. yel.	Nor. yel.	Str. yel.	Nor. yel.	Str. yel.	Nor. yel.	Nor. wh.	Str. yel.	Str. yel.	Str. wh.	Nor. yel.	Nor. wh.	Str. yel.	Str. wh.	Str. yel.	Str. wh.	Nor. yel.	Nor. wh.	Str. yel.	Str. wh.	Nor. yel.	Nor. wh.					
	194	80	100	86	35	210	80	192	91	81	459	204	23	12	54	155	61	247	26	24	65	60	3	10	18					



Striped yellow ♀ × Striped yellow ♂
(S.3'10)

1910	Striped yellow ♀ × Striped yellow ♂ (S. 3'10)									
	├── Str. yel. Pl. yel. (S. 5'11)									
	129	60								
	♀ × Normal white (<i>Aojiku</i> , brown-anted) ♂									
	├── Str. yel. Nor. wh. (H. 31'12)									
	215	188								
	├── (1)	(2)	(3)	(4)						
	Str. yel. Str. wh. Nor. yel. Nor. wh. Str. yel. Nor. wh. Str. yel. Nor. wh. Str. yel. Nor. wh.									
	204	29	32	54	154	157	219	229	209	194
1913										

Among the four families derived from II. 31'12, the result in (1) is explained on the basis of the usual system of reduplication of **S-Y** coupling¹, the results in the rest show, on the contrary, quite a distinct feature. In these latter families we find only two zygotic forms in nearly equal ratio, the actual numbers in total being

	Str. yel.	Nor. wh.
Nos. 2, 3, 4 all together	582	580

We cannot consider above results as due to a complete coupling of an ordinary nature, because we should expect, if such were the case, double dominant and double recessive forms in the ratio 3 : 1 instead of 1 : 1 ratio which was attained by the present families.

If the simultaneous dropping out of the factors **S** and **Y** in the male parent has been assumed, above results may be simply explained. But we have at present no positive evidence in support of this assumption.

4. ON THE GENETIC CONSTITUTIONS OF THE STRIPED AND MORICAUD MARKINGS.

The question dealt with here concerns whether the genetic constitutions of the striped and moricaud markings (in homozygous condition) are **SSnn** and **MMnn** respectively, or are actually **SSNN** and **MMNN**.

As experimental results described elsewhere show, F_2 families ex striped \times normal involved only striped and normal individuals, but no other marking, plain for instance. On the basis of **SSnn** view, on the other hand, we are to expect three forms of marking in F_2 , i. e. striped, normal and **ssnn** markings. As this did not actually happen, however, we must, for explanation of the phenomenon, assume a complete repulsion between **S** and **N**.

If the formula **SSNN** was accepted, on the contrary, above result is simply analysed without need of assuming any reduplication, but another difficulty occurs here. In F_2 ex **SSNN** \times **ssnn** (plain) we should expect

1. (2 : 1 : 1 : 2) ♂ \times (1 : 1) ♀. (cf. p. 5.

the normal-marked ones besides the striped and plain-marked specimens. This is contrary to the actual results, F_2 generation practically involving striped and plains but no individuals normally marked. Thus we are forced again to assume a reduplication, namely, complete coupling between **S** and **N**.

The same thing may be said as regards the relation of the formulae **MMnn** and **MMNN** i. e. we must assume complete repulsion between **M** and **N** for **MMnn** system, and complete coupling between them for **MMNN** view.

On the striped and moricaud larvae the normal marking ("eye-spots" and "lunules") co-exists with that marking which is characteristic of the striped or the moricaud. This fact seemingly points to the **SSNN** and **MMNN** formulae. But it is, at the same time, not unreasonable to suppose that the normalness actually partakes of the striped or the moricaud marking, the characteristic being freely developed by **S**- or **M**-factor in absence of **N** as well as in presence of it.

We are, as these comparisons show, unable to determine, at present, which of the formulae above stated are more adequate. Therefore I will prefer, until more positive evidence to the contrary is obtained, the expressions **SSnn** and **MMnn** which were adopted in my previous papers.

February 1914.

Agricultural College,

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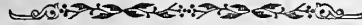
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APR 29 1924

ON THE OCCURRENCE OF CREATININ IN LEGUMINOUS SEEDS.

By

K. Oshima and M. Ariizumi.

INTRODUCTORY.

Creatinin is found in muscles and bloods of animals in varying quantities, accompanying creatin, though the former in much smaller quantity than the latter. In living muscles of many fishes it is present in a comparatively large amount. Together with creatin it forms one of the normal constituents of urine of mammals.

Concerning the origin of creatinin and its relation to creatin various views have been advanced. But it is a well established fact that creatin when administered either per os, or sub cutan, a part of it at least, is excreted in urine, as creatinin. Creatinin in food is resorbed from the intestinal walls and is excreted again, largely as such, in urine. Aside from these two sources, creatinin seems to be produced as the result of metabolism in animal bodies, with creatin as its intermediate form. While its physiological rôle in animal metabolism is not yet clearly understood, its sources have generally been considered as wholly confined to animal origin.

Though, the literature concerning the biochemistry of the animal organism is filled with references to creatinin, the possibility of its occurrence outside of the animal kingdom has not been investigated to any great

extent. Zinno¹⁾ found that cultures of a certain bacteria built creatinin in the nutrient medium. Antonoff²⁾ also reported the formation of creatinin by bacteria in pepton solution. Recently, E. C. Shorey³⁾ found it in the alkaline as well as alcoholic extracts of soils. He maintained that the considerable portion of the isolated creatinin has been present in the soil as such. According to him, nucleic acid and phytin in soils have some relation with the formation of creatinin. Shortly after the Shorey's study, M. X. Sullivan⁴⁾ has found it in the vegetable kingdom for the first time. He determined creatinin in wheat seeds, wheat seedlings and wheat bran, in rye, clover and alfalfa seeds, in mature cowpea plants and in potato tubers. He found also that both the planted soil and the soil which had not recently been cropped, contained creatinin, but it was present in larger amounts in the recently cropped soil. From this point of view, he asserted that the presence of creatinin in the soil is connected in some way with plant growth. He reported that creatinin seems to persist for a considerable time in soil and may increase in it by accumulation, and its presence in plants and in the medium in which plants grow has considerable bearing on soil fertility. J. J. Skinner⁵⁾ has shown that creatinin has a marked beneficial effect on plant growth by culture experiment. Under the direction of one of the authors K. Ebiko⁶⁾ determined the presence of creatinin in *Lespedeza bicolor*, Turcz., var. *Typica*, Max., which is used in Japan for fodder as well as for green manure.

In the following pages are presented the results of our experiments concerning the presence of creatinin in several leguminous seeds, which are generally consumed by Japanese as food stuffs. In all the legumes we have studied, the presence of creatinin was fully ascertained.

-
- 1). *Reforma med.*, Roma. **9**, (1893) (3), p. 806; through *Jour. Amer. Chem. Soc.*, **33**, (1911), p. 2036.
 - 2). *Centrabl. Bakt.*, Jena, **43**, (1907), p. 209.
 - 3). *Science*, **33**, (1911), p. 340.
 - 4). *Jour. Amer. Chem. Soc.*, Easton, Pa. **33**, (1911), pp. 2035-2042.
 - 5). *Bot., Gaz.* **54**, (1912), pp. 152-163.
 - 6). *Jour. Soc. Agr. & Forest.*, Sapporo, No. 25, 1914.

EXPERIMENTAL.

As the materials for the present investigation the following leguminous seeds were selected.

- | | |
|---------------------|--|
| 1. Adzuki bean. | <i>Phaseolus radiatus</i> var. <i>aurca</i> Prain. |
| 2. Black soy bean. | <i>Glycine hispida</i> Maxim. |
| 3. Yellow soy bean. | „ |
| 4. Kidney bean. | <i>Phaseolus vulgaris</i> L. |
| 5. Green peas. | <i>Pisum sativum</i> L. |
| 6. Horse bean. | <i>Vicia Faba</i> L. |

The seeds were washed with water to remove dusts and other impurities, dried and ground in the mill as usual. To remove fats and oils contained in the seeds, about double volumes of ether were added to the ground preparation and allowed to stand a day under frequent shaking. The clear ether extract was then decanted and filtered. To the residue another portion of ether was added and after standing for a day under frequent shaking, it was filtered. The residue was then left at the room temperature until the smell of ether was no more appreciable. As the soy beans contained such a large amount of oil that the above process was not found satisfactory, they were extracted in the Soxhlet's apparatus with ether in the usual manner.

The sample thus prepared was boiled in a large flask provided with reflux condenser, with strong alcohol, for several hours. The alcoholic extract was filtered hot, and the residue was pressed to remove the extract well. The extract thus obtained was allowed to stand 24 hours, when some precipitates were found to be formed. They were separated by filtration and the clear filtrate thus obtained was concentrated by evaporating in a partial vacuum. As the solution became concentrated, some fatty substances seemed to separate out, the liquid was transferred to a porcelain basin and stirred with a glass rod under the addition of water, when fat-like substances adhered to the bottom of the basin as well as to the glass rod. The clear solution was decanted and evaporated on a

water bath at a low temperature, under constant stirring, until a syrupy solution was obtained. The syrup thus prepared was used for the identification of creatinin and is designated as syrup I in the statement beyond.

Color Reactions.

The following three reactions were made use of for the determination of creatinin.

a) *Jaffé's reaction.* An intensive red color is produced when to the aqueous solution of creatinin are added, first, a little picric acid and, then, a few drops of caustic alkali. The red color disappears on adding excess of alkali or on acidifying the solution with acetic acid or with hydrochloric acid. The red color appears soon after the addition of caustic alkali even in cold. The intensity varies from orange red to dark blood red according to the amount of creatinin present. The characteristic red color will soon change into yellow if too much alkali is used and particularly when exposed to light. The presence of glucose does not interfere with this reaction, since the similar red color produced by glucose itself appears much more slowly than in the case of creatinin. It should be remembered in this connection that acetone, laevulinic acid and furfural give the *Jaffé's* reaction.

b) *Weyl's reaction.* When a freshly prepared, very dilute solution of sodium nitroprusside is added to the aqueous solution of creatinin until it gives a distinct yellow color, and then a few drops of dilute caustic alkali are added, the solution gives a ruby red color. The color becomes lighter as the time passes on until after a short time it assumes a straw-yellow color. Salkowski¹⁾ asserted that ruby red color can not be considered as characteristic because this color easily changes into yellow. In the course of our investigation we have observed the same fact, in confirmation of Salkowski's view. Furfural gives the same color with sodium nitroprusside and caustic alkali as in the case of creatinin, the only difference being that the red color given by furfural remains unchanged for a much longer time. Laevulinic acid gives a color similar to that of creatinin. Sulphide gives purple color under the same conditions and makes the creatinin reaction obscure if both are present at the same time.

c) *Salkowski's reaction.* If the yellow solution obtained in testing *Weyl's* reaction, is acidified with acetic acid and heated, it turns first green, then blue and finally the precipitate of Prussian blue is separated. Hydantoin gives the same reaction but not creatinin. Laevulinic acid and furfural give the same final blue color and precipitation, but the solution is turned purple directly after acidifying with acetic acid and before heating. The reagents alone will give the same final color and precipitation if heated before acidifying.

1) Hoppe-Seyler's Zs. Physiol. Chem., Strassburg, **4**, (1880), p. 133.

A) About 1 cc. of the syrup I was taken and the color reactions were tested, according to the methods already described. The results obtained are shown in the following table.

Table I.

Syrup from	Reducing sugar	Jaffé's reaction		Weyl's reaction		Salkowski's reaction	
Adzuki bean	—	+	+	+	+	+	+
Black soy bean	—	+	+	+	+	+	+
Yellow soy bean	—	+	+	+	+	+	+
Kidney bean	+	+	+	+	+	+	+
Green peas	—	+		+		+	
Horse bean	—	+		+		+	

B) The syrup I was diluted somewhat with water and then neutral lead acetate was carefully added, until no more precipitate was formed and after allowing to stand 24 hours it was filtered. The excess of the lead in the filtrate was removed by hydrogen sulphide. The lead-free filtrate was concentrated to a syrup (syrup II) under the reduced pressure. The results were as follows:

Table II.

Syrup from	Jaffé's reaction		Weyl's reaction		Salkowski's reaction	
Adzuki bean	+	+	+	+	+	+
Black soy bean	+		+		+	+
Yellow soy bean	+		+		+	+
Kidney bean	+		+		+	+
Green peas	+		+		+	
Horse bean	+		+		+	

Isolation of Creatinin as Creatinin Zinc Chloride.

If the solution under examination contains compounds giving similar color reactions as creatinin, the color reaction alone can not be depended upon as the criterium for the presence of creatinin in it. Hence, when the indications for the presence of creatinin were obtained by the color tests, it was confirmed and established by the preparation of the characteristic crystals of creatinin zinc chloride. Creatinin zinc chloride forms prisms or slender needles, grouped in rosettes, clusters and stars. The crystal is formed when concentrated solutions of creatinin and of zinc chloride are mixed in the absence of free mineral acids, a condition usually obtained by the addition of a little sodium acetate. When a large quantity of creatinin is treated in the manner above described there is immediate precipitation, but when small quantities are being treated, the precipitation does not begin for several hours and not complete for several days. The time required for crystal-formation seems also to be greatly influenced by the amount of impurities present in the syrup.

A. From impure syrup.

To the syrup II the solution of sodium acetate and of zinc chloride were added and the mixture was kept in a desiccator over concentrated sulphuric acid. Gradually the solution became concentrated and the formation of characteristic crystals of creatinin zinc chloride was observed, but not in all cases. The time required for crystal-formation, the forms of crystals etc. are described in the following.

Adzuki bean. The characteristic crystals appeared after 10 days in the forms of stars, crossed needles and clusters, and after 16 days they grew larger and well developed monoclinic plates were observed.

Black soy bean. Crystals were not observed.

Yellow soy bean. „

Kidney bean. The characteristic crystals appeared after about 3 weeks. They were radiated needles or crosses.

Green peas. The characteristic crystals appeared after about 10 days. Crystal forms were the same as in the case of kidney bean.

Horse bean. The characteristic crystals appeared after about 3 weeks in the form of radiated needles or crosses.

B. From purified syrup.

The syrup II seemed to contain still some impurities which prevented more or less the crystallisation of creatinin zinc chloride. The syrups obtained from both kinds of soy bean which showed apparently strong color reactions of creatinin did not show any sign of forming characteristic crystals of creatinin zinc chloride under the similar conditions in which the syrups from other legumes formed the crystals. The syrups from soy beans evidently contained some impurities which had preventive influence on the formation of the crystal. Starting from this point of view we have attempted first to separate creatinin from admixed substances and then to try the isolation of creatinin as its zinc chloride salt. For the separation of creatinin from the syrups of leguminous seeds, we have adopted the following procedure.

The alcoholic extract of the seed was concentrated to a small volume under reduced pressure and treated with neutral lead acetate to remove impurities. The excess of lead in the filtrate was removed by hydrogen sulphide. The lead-free filtrate was concentrated to about 30 cc. and a small quantity of glucose was added. The hot solution thus prepared was then poured into 50 cc. of the boiling Fehling's solution. The solution was kept boiling for 2 minutes and then allowed to cool, when a greyish white gelatinous precipitate of creatinin cuprous oxide, mixed with brownish red precipitate of cuprous oxide was separated. The precipitate formed was filtered, well washed with 95 % alcohol, suspended in water and decomposed with hydrogen sulphide. The copper sulphide was separated by filtration and the filtrate was concentrated to a small volume under reduced pressure and, with a small portion of it, color reactions were tested, after the complete removal of the hydrogen sulphide by careful evaporation, since its presence makes the Jaffé's and Weyl's reactions obscure. The results are shown in the following table.

Table III.

Syrup from	Jaffé's reaction	Weyl's reaction	Salkowski's reaction
Adzuki bean.	+	+ +	+ +
Black soy bean.	+ +	+ +	+ +
Yellow soy bean.	+	+	+ +
Kidney Bean.	+	+	+
Green peas.	+ +	+ +	+ +
Horse bean.	+ +	+ +	+ +

From the results obtained, it is seen that in all cases the creatinin was precipitated with cuprous oxide and recovered again by hydrogen sulphide.

To the concentrated solution obtained in the above treatment, saturated solution of zinc chloride and a little sodium acetate were added and the whole was allowed to stand several days. Within a few hours crystals began to form, and in a few days they were observed to have the characteristic appearance of creatinin zinc chloride. The time required for crystal-formation and the forms of crystals are shown in the following.

Adzuki bean, black soy bean and yellow soy bean. Crystallisation completed in 24 hours. Crystal-forms were radiating needles, crosses and clusters.

Kidney bean. Crystallisation completed in 24 hours. Crystal-forms were radiating needles, crosses, clusters and plates.

Green peas. Crystallisation completed in 48 hours. Crystal-forms were stars, rosettes and plates.

Horse bean. Crystallisation completed in 5 days. Crystal-forms were radiating needles, crosses and clusters.

It is thus seen that by separating creatinin with cuprous oxide from the admixed impurities, the crystallisation could be effected much sooner than in the case of direct treatment. Even in the extract of soy bean with which we were unable to obtain the characteristic crystals of creatinin zinc chloride by direct treatment, the crystallisation was completed within 24 hours. In every case, the crystal-forms were very distinct and free

from impurities when seen under the microscope.

Regaining of Creatinin.

The regaining of creatinin from creatinin zinc chloride was undertaken in the following manner.

The crystals obtained from the extract of Adzuki bean were separated from the mother-liquor by filtration and well washed with strong alcohol. The residue on the filter was dissolved in boiling water and filtered hot and the filtrate was allowed to recrystallise after concentration. The operation was repeated once more. The crystals thus obtained were white in color and when observed under microscope, they were somewhat oblong hexonal plates. They were dissolved in water and boiled with some freshly prepared lead hydroxide, filtered, and the filtrate was concentrated to a small volume. The concentrated solution gave Jaffé's reaction so distinctly that there is no doubt of the presence of creatinin in it. Unfortunately from the lack of the material, other reactions could not be observed.

The crystals of creatinin zinc chloride from the extract of legumes other than Adzuki bean were so little that they did not allow further working up.

Summary.

1) The color tests, the formation of characteristic double salt of creatinin zinc chloride and lastly, the regaining of creatinin from the double salt, were applied to determine the presence of creatinin in legumes.

2) The presence of creatinin was confirmed for the first time, in the seeds of Adzuki bean, black soy bean, yellow soy bean, kidney bean, horse bean and green peas.

3) The amounts of creatinin in the seeds of Adzuki bean, kidney bean and soy beans are apparently in much larger quantity than in horse bean and green peas, though, its absolute amount seems to be very small.

(March, 1914)



ÜBER DIE KOLLOIDALEN EIGENSCHAFTEN DER SAUREN BÖDEN IN JAPAN

von

T. Tadokoro, *Nōgakushi.*

In jüngerer Zeit wurden von verschiedenen Autoren kolloid-chemische Arbeiten am Boden vorgenommen, und viele beachtenswerte Untersuchungsergebnisse geben uns interessante wissenschaftliche Aufschlüsse. Über die kolloidalen Eigenschaften der in Europa sehr ausgedehnten humusreichen sauren Böden wurden in neuester Zeit viele Berichte erstattet, und die Bedeutung der Kolloide für die saure Eigenschaft einigermaßen erklärt. Mit humusarmen, mineralischen sauren Bodenarten wurden von amerikanischen und japanischen Forschern verschiedene Versuche unternommen. Besonders Prof. Dr. G. Daikuhara¹⁾ hat sich mit dem Ursprung, dem Nachweis und der Bestimmungsmethode der Acidität des mineralischen sauren Bodens in Japan befasst. Aber bis jetzt sind von ihm noch keine Untersuchungsergebnisse über kolloidale Eigenschaften mitgeteilt worden.

Über die kolloiden Substanzen im Boden: Zunächst haben wir zwischen festen kolloiden Substanzen und kolloiden Lösungen zu unterscheiden. Erstere sind amorphe Körper mit geringer Kohäsion und letztere, von weit grösserer Wichtigkeit, die mit einem Lösungsmittel extrahierten. Um die allgemeine Beschaffenheit der kolloiden Substanzen im Boden zu erkennen, haben wir zunächst die folgenden Versuche angestellt, die sich auf die erste Gruppe beziehen. Über die kolloiden Lösungen wollen wir in nächster Zeit ausführlichen Bericht erstatten. Die verschiedenartigen

1) Chem. Zeitug., (1908), Nr. 98

Bulletin Imp. Centr. Agric. Exp. Station, Japan, (1914), II, 1.

[Jour. of the College of Agr., Tohoku Imp. Univ., Sapporo, Vol. VI, Pt. 2, June, 1914.]

Proben der humusarmen sauren Böden, die in meinen Versuchen verwendet wurden, hat der verstorbene Prof. Dr. S. Suzuki im letzten Jahre auf weiten Gebieten Japans gesammelt.

Wahl der Proben.

Bei der Entnahme von saurem Boden zu unsern Versuchen wurden nachfolgende 12 Proben ausgewählt. Die frischen Proben wurden an der Luft vorgetrocknet und durchgesiebt. (Dia. 0,5 mm)

Nummer der Proben	Wasser gehalt in %	Acidität n. d. Daikuha-ra-KCl- methode (ccm.)	Sammelort.	採 集 地
1	2,85	23,88	Fukuiken Matsudaira Landw. Versuchsstation.	福井縣松平農事試驗 場
2	3,00	46,68	Niigataken Landw. Versuchs- station.	新潟縣農事試驗場
3	5,00	171,10	Shizuokaken, Shidagun.	靜岡縣志太郡岡部町 岡部字札澤
4	4,59	70,63	Shizuokaken, Hikusagun.	靜岡縣引佐郡西濱名 村日比澤字駄荷野
5	3,76	61,49	Shigaken, Kogagun.	滋賀縣甲賀郡大野村 字大野
6	2,88	45,07	Shigaken, Kogagun.	同 上 水口町 大字水口
7	6,78	43,26	Hokkaidō, Iburinokuni.	北海道膽振國虻田郡倶知 安村基線通西八號
8	8,50	44,36	„ „	同 上
9	5,99	73,55	Naraken, Yoshinogun.	奈良縣吉野郡吉野村 字丹治
10	13,38	323,60	Ishikawaken, Hasakugun.	石川縣羽咋郡比色知 村字神子原
11	5,34	90,55	Ishikawaken, Ishikawagun.	石川縣石川郡弓取村 字安江
12	4,95	79,02	Ishikawaken, Kahokugun.	同 上 河北郡花園村 字朝日

Versuch 1. Quellung.

Ein Kolloid vergrößert sein Volumen bei der Berührung mit Wasser, es quillt. Die Quellung ist eine physikalisch-chemische Eigenschaft der

Gallerten, der hydrophyilen Kolloide, besonders organischen Ursprungs. Eine ähnliche Regel dürfte für Laugen gelten, und zwar ist die Quellung in Alkalien stärker als die in Salzlösungen. Die kolloiden Substanzen organischen Ursprungs, wie Eiweiss und Stärkearten, haben viele Autoren wie J. Reinke¹⁾, F. Hofmeister²⁾, E. Rodewald³⁾, Wo. Pauri⁴⁾, K. Spiro⁵⁾, E. Overton⁶⁾, W. Ostwald⁷⁾, H. Fischer⁸⁾, u. s. w. untersucht.

Quellungserscheinungen wurden ebenfalls bei den Bodenkolloiden bemerkt, wie K. Stremme⁹⁾, der die Allophatone in Mineralböden untersuchte, nachgewiesen hat. Daher haben wir mit einigen chemischen Lösungen die verschiedenen Bodenarten geprüft und nachfolgende Resultate gefunden.

Zur Prüfung des Quellungsvermögens dieser Proben wurde ein Gemisch von 5 g des Bodens mit 15 ccm der Reagenzien 30 Minuten lang geschüttelt und nach 24 Stunden das vergrösserte Volumen bestimmt.

Tabelle 1.

Nummer der Proben	Quellung mit Alko hol	Quellung mit Wasser	mit 5 % Na Phos- phatlösung	mit 5 % Na Acetat- lösung	mit 5 % Na Tar- taratlösung	mit 5 % Na Car- bonatlösung	mit 5 % NaOH	mit 5 % HCL	mit 5 % Essigsäure
1	5,3	5,7	5,3	5,1	5,3	5,4	5,4	5,7	5,7
2	5,1	5,0	5,4	4,9	5,1	5,0	6,1	5,4	5,2
3	5,6	8,2	6,1	5,9	7,5	7,0	7,1	7,1	6,5
4	6,15	—	6,9	6,4	6,8	7,2	9,0	7,5	6,7
5	5,9	6,3	6,2	5,5	6,0	6,0	7,1	6,5	5,8

- 1) Hansteins botan. Abhandlungen., 4, (1879), 1.
- 2) Arch. Exp. Path. u. Pharm., 27, (1890), 395 : 28, (1891), 210.
- 3) Zs Physik. Chem., 24, (1897), 193.
- 4) Pflügers Arch., 67, (1897), 225.
- 5) Beitr. Chem. Physiol., 5, (1904), 276.
- 6) Pflügers Arch., 105, (1904), 176.
- 7) Ibid, 106, (1905), 568.
- 8) Das Oedem im experim. u. therapeut. Unterricht der Physiologie u. Pathologie d. Wasserbindungen in Organismen., Dresden, (1910).
- 9) Ber. d. Deutsch. geol. Ges., 122, (1010), 128.

6	5,8	6,1	5,6	5,6	5,7	5,9	6,8	6,0	5,5
7	7,6	7,7	8,0	8,0	8,3	9,4	13,2	8,7	8,6
8	7,65	8,6	9,3	8,5	9,2	12,1	15,8	10,5	8,6
9	6,15	7,5	6,1	6,3	6,8	7,1	8,4	7,3	7,6
10	—	—	—	—	—	—	—	—	—
11	6,9	7,8	6,3	9,0	6,4	7,0	8,0	7,2	6,5
12	5,4	5,7	5,8	5,6	5,7	6,3	7,0	6,4	5,9

Prozentzahlen der vergrößerten Volumen der geglähten Proben:

1	8,18	16,13	8,16	4,08	8,16	12,24	12,24	18,29	18,29
2	6,25	4,17	13,50	2,08	6,25	4,16	27,08	13,50	6,25
3	0	28,74	8,93	5,35	33,92	26,78	33,92	26,56	16,07
4	5,17	—	19,00	12,07	17,24	22,41	55,17	29,31	15,50
5	22,22	28,57	10,20	12,24	22,44	22,44	44,89	30,61	18,36
6	1,37	19,60	7,84	7,84	9,80	15,69	33,33	17,64	7,84
7	28,66	28,38	33,33	33,33	38,33	83,33	85,20	45,00	43,33
8	22,50	36,50	47,73	34,87	46,03	92,22	136,50	66,66	36,50
9	0,83	37,70	49,18	36,06	47,54	95,08	140,98	68,85	37,70
11	15,00	8,33	10,00	50,00	6,66	16,66	33,33	16,66	8,33
12	0	5,55	7,40	3,73	5,55	16,66	42,22	22,22	9,26

Die obigen Werte, sowie die vergrößerten Volumen, wurden in den Vergleichen aus folgenden geglähten Proben berechnet.

Tabelle 2.

Nummer der Proben	1	2	3	4	5	6	7	8	9	11	12
Quellungs- volum in ccm	4,9	4,8	5,6	5,8	4,9	5,1	6,0	6,3	6,1	6,0	5,4

Zur Erklärung der Beziehungen der kolloiden Substanzen, die aus dem Boden mit Wasser ausgelaugt wurden, wurde der folgende Versuch zur Bestimmung des Quellungsgrades des Bodens gemacht. Die mit

Wasser extrahierten Böden wurden filtriert und mit Wasser gewaschen. Die auf dem Filter gebliebenen Reste dienten im Quellungsversuch als Proben. Die Gemische von 5 g des Bodenrestes und 15 ccm Wasser wurden 30 Minuten geschüttelt und die vergrößerten Volumen beobachtet.

Tabelle 3.

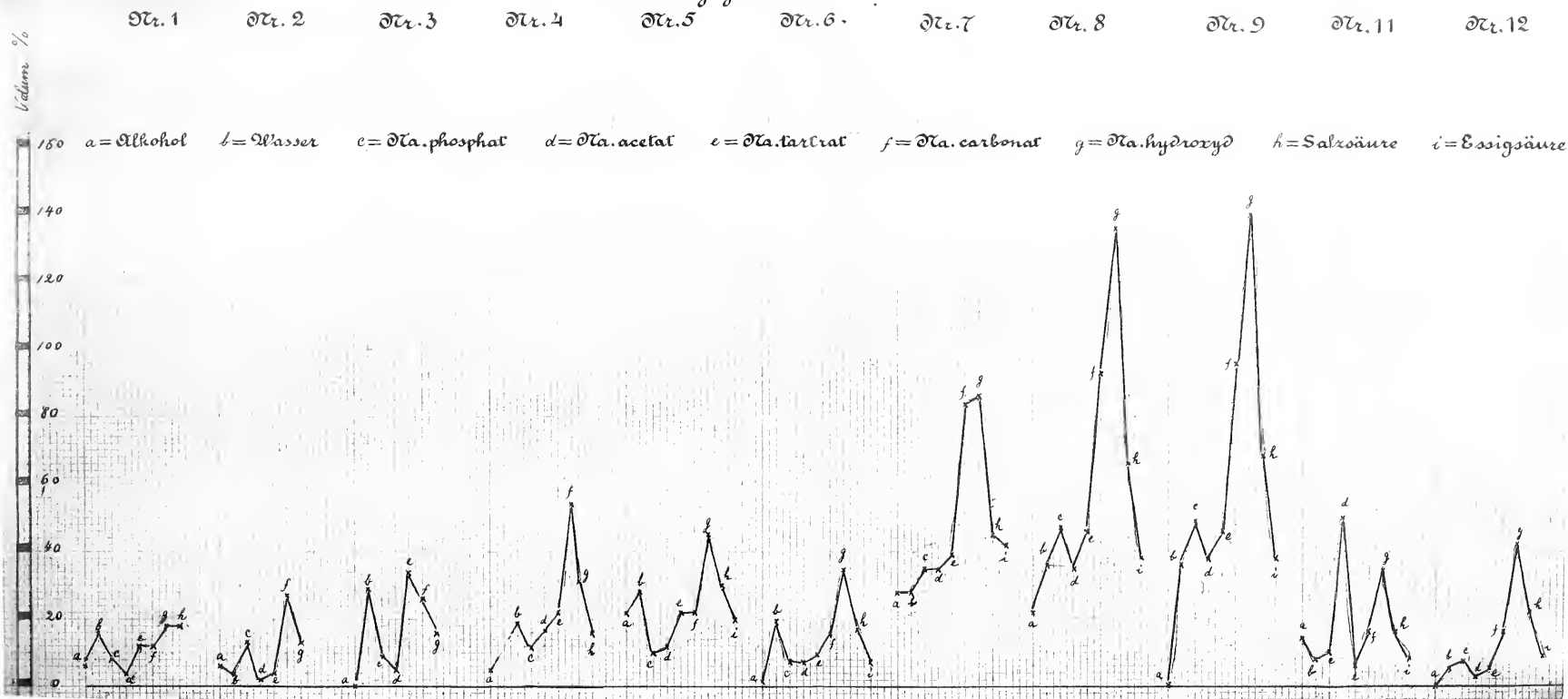
Nummer der Proben	1	2	3	4	5	6	7	8	9	11	12
Quellungs- volumen.	5,6	5,0	7,0	—	6,3	5,9	7,8	8,7	7,3	6,7	6,0
Volumen, d. oben beschr. Proben.	5,7	5,0	8,2	—	6,3	6,1	7,7	8,6	7,5	6,5	5,7
Unterschied.	—,1	0	—,1	—	0	—,2	—,1	—,1	—,2	—,2	—,3

Wenn in der nebenstehenden Figur die vergrößerten Volumen auf der y Achse und die verschiedenen Reagentien auf der x Achse aufgetragen werden, so kann man aus jeder Kurve aus ihrer Entfernung von der x Achse die Werte leicht ersehen.

Der Boden hat viele Arten von Gel-bildungssubstanzen, die mit Flüssigkeiten eine Adsorptionsverbindung bilden und daher Aufquellungserscheinungen zeigen. An diesen Erscheinungen erkennen wir die Gelform und den Bau des Gels im Boden. Wie man aus der obigen Figur ersieht, sind die Quellungsgrade desselben Bodens je nach der Art des Reagens veränderlich, ein bestimmter Boden gibt ferner keine Anhaltspunkte für die Vergrößerung des Volumens anderer Böden. Diese Tatsache zeigt uns, dass die gebildeten Gelformen nach der Art des Reagens verschieden und die Gelbildungsmaterialien nicht einheitliche Substanzen sind.

Die Unterschiede der vergrößerten Volumen der originalen Proben und der dialysierten Reste sind sehr klein. Daher haben die kolloiden Substanzen, die aus dem Boden mit Wasser ausgelaugt wurden, keine wichtige Wirkung auf die Quellung des Bodens.

Quellungsgrade





Versuch 2. Quellungswärme.

Quellungsvorgänge sind stets von Wärmeentwicklung begleitet. So fanden E. Wiedemann und Ch. Lüdeking¹⁾ bei der Quellung von Gelatine, Tragant und Gummi arabicum Temperaturerhöhungen bis zu 1,9°. Ja W. Hardy²⁾ gibt sogar an, dass beim Zusammenbringen von 1 ccm trocknen Agars mit 10 ccm Wasser Temperaturerhöhungen von mehr als 6° zu beobachten sind. Die Benetzungswärme des Bodens wurde schon von vielen Autoren bemerkt und beträgt 0,5—1,0 Kalorie für Sandboden und 4,5—10,0 Kalorien für Humusboden.

In Gegenwart von löslichen Salzen im Boden gibt es keine Quellungswärme, und da der Gehalt an gelösten Stoffen bei jeder Probe verschieden ist, so ist sie schwer messbar.

Über die Abhängigkeit der Quellungswärme vom Wassergehalt hat Rodewald³⁾ berichtet, dass die entwickelte Wärmemenge mit sinkendem Wassergehalt wächst, und dass die grösste Wärmemenge bei der Aufnahme der ersten Menge Wasser frei wird.

Bei unsern Versuchen wurden zunächst die Proben in der Dialyse ausgesalzt, dann im Wasserbade getrocknet, gepulvert und hierauf die Quellungswärme bestimmt.

Für diese Wärmemessungen wurde das Wasseräquivalent eines Kalorimeters nach der Neutralisationswärme von 1/2 Normalschwefelsäurelösung und 1/2 Normalnatriumhydroxydlösung bestimmt. Hierauf nahmen wir für den Wert der Neutralisationswärme 147,5 Kalorien und bestimmten nach folgender Gleichung das Wasseräquivalent. Es ergab 23,2.

$$t = \frac{m_1 t_1 + m_2 t_2}{m_1 + m_2}, \quad \left\{ (v_1 \times d_1 + v_2 \times d_2) C + M \right\} (T - t) = 147,5$$

$M = 23,2$

„t“ bedeutet die anfängliche Temperatur des Gemisches, „t₁“ die Temperatur, „m₁“ das Mass, „v₁“ das Volumen und „d₁“ die Dichte der 1/2 N. Schwefelsäurelösung; „t₂“ die Temperatur, „m₂“ das Mass, „v₂“ das Volumen und „d₂“ die Dichte der 1/2 N. Natriumhydroxyd-

1) Bechhold, H.—Die Kolloide in Biologie u. Medizin., (1912), 122, 126.

2) Zs. f. physik. Chem., 33, (1900), 326.

3) Ibid., 24, (1897) 246.

lösung. „C“ die spezifische Wärme, „T“ die endliche Temperatur des Gemisches. „M“ ist das gesuchte Wasseräquivalent.

Die Bestimmung der Quellungswärme wurde ausgeführt an 50 g trockenem Boden und 500 g Wasser; die Temperaturänderungen wurden mit einem Beckmannschen Differential Thermometer in jeder Minute gemessen.

Die Wärmemengen, die beim Aufquellen frei werden, wurden nach den oben beschriebenen Gleichungen berechnet und folgende Resultate gewonnen.

Tabelle 4.

Nummer der Proben	1	2	3	4	5	6		8	9	10	11
Kalorie	1,65	1,76	2,95	—	3,01	2,44	2,97	6,66	3,01	—	1,80

Die Zahlen zeigen, dass die Quellungstemperatur in der Probe 8 am höchsten steigt, und die Quellungswärme ist ca. dreimal so gross wie die von Probe 1.

Vergleichung der Quellungsgrade mit der Quellungswärme.

Tabelle 5.

Nummer der Proben	1	2	3	4	5	6	7	8	9	10	11
Quellungs-wärme	1,65	1,76	2,95	—	3,01	2,44	2,97	6,66	3,01	—	1,82
Quellungs-volumen	5,6	5,0	2,9	—	6,3	5,9	7,8	8,7	—	6,7	6,0
Verhältnisse Q. V./Q. W.	3,3	2,8	2,4	—	2,3	2,4	2,6	1,3?	—	—	3,2

Die Werte dieser Vergleichen zeigen gewöhnlich regelmässige Verhältnisse (ausser der Probe 8, die an Humussäuregehalt¹⁾ reich ist).

1) Die freien Humussäuremengen sind wie folgt. (% der Trockensubstanzen)

Proben	1	2	3	4	5	6	7	8	9	10	11	12
%	1,139	1,174	2,15	3,83	3,71	2,64	5,49	10,18	1,50	—	2,45	1,92

Daraus ersehen wir, dass die Quellungswärme dem Quellungsgrade in humusarmem saurem Boden proportional ist.

Versuch 3. Hygroskopizität.

Die Hygroskopizität ist die absorptive Bindung des Wasserdampfes durch Oberflächen und für das Studium des Kolloidgehalts der Böden bedeutungsvoll. Die Schwächen der Hygroskopizitätsbestimmung, sowie ihre grossen Vorzüge und überhaupt ihre Eignung für die Beurteilung von Böden sind von P. Vageler¹⁾ vom kolloidchemischen Standpunkt aus diskutiert worden. Die Hygroskopizität ist die Wassermenge, welche der Boden enthält, wenn seine Oberfläche mit einer Molekülschicht Wasser bedeckt ist, bezogen auf hundert Gewichtsteile des trocknen Bodens. Die Hygroskopizität ist also eine der Bodenoberfläche proportionale Grösse; die Menge des hygroskopisch gebundenen Wassers schwankt für die verschiedenen Bodenarten in sehr weiten Grenzen. Mit dem Gehalt an kolloidem Ton und an Humus wächst die Menge des hygroskopischen Wassers.

Um die Beziehungen der Hygroskopizität zum Kolloidgehalt zu erkennen, unternahmen wir folgenden Versuch. Wobei die Bestimmung der Hygroskopizität erfolgte nach dem Verfahren von H. Rodewald und A. Mitscherlich. Das Verfahren besteht aus folgenden Grundzügen: es wird zunächst der lufttrockene Boden in einem mit Phosphorpentoxyd versehenen Exsikkator behandelt und dann in den 10 % ige Schwefelsäure enthaltenden Apparat gestellt und die aufgenommene Wassermenge bestimmt. (Vergl. J. König: Unters. landw. Gewerb. Stoffe. vierte Aufl. S. 67.)

Tabelle 6.

Proben	Hygroskopizität	Proben	Hygroskopizität	Proben	Hygroskopizität
1	3,40	5	4,48	9	6,92
2	3,26	6	3,54	10	—
3	7,13	7	7,95	11	5,42
4	5,98	8	10,02	12	3,82

1) Fühlings Landw. Ztg. 61, (1912). 73.

Aus diesen Zahlen ergibt sich, dass die Hygroskopizität in der Probe 8 am höchsten und in der Probe 2 am geringsten ist. Die durchschnittliche Zahl beträgt 5,62.

Vergleichung der Quellungsgrade mit der Hygroskopizität.

Tabelle 7.

Proben	Hygroskopizität = Hy.	Quellungsvolumen = Qv.	Verhältnisse $\frac{Qv.}{Hy.}$
1	3,40	5,7	1,6
2	3,26	5,0	1,5
3	7,13	8,2	1,2
4	5,98	—	—
5	4,48	6,3	1,4
6	3,54	6,1	1,7
7	7,95	7,7	0,98?
8	10,02	8,6	0,86?
9	6,92	—	—
11	5,42	6,5	1,2
12	3,82	5,7	1,5

Diese Vergleichen zeigen, dass die Verhältnisse einander sehr nahe kommen, ausser den Proben 7 u. 8, die an Humussäure reich sind (l. c.). Wir erkennen, dass die Hygroskopizität gewöhnlich in nahen Beziehungen zum Quellungsgrade steht.

Versuch 4. Adsorption.

Die Bestimmung der Kolloide durch Bestimmung der Adsorptionsfähigkeit des Kolloides für Farbstoffe (Fuchsin, Methylenblau und Methylviolet) und Salzlösungen schliesst sich an die Versuche von F. Cornu¹⁾, K. Endell²⁾ und B. Sjollem³⁾. Mehrere andere Autoren bestimmen die

1) Koll. Zs., **4**, (1909), 304.

2) Ibid., **4**, (1909), 246.

3) Journ. f. Landw., **53**, (1905), 67, **53**, (1905), 70.

Adsorptionsfähigkeit der verschiedenen Böden durch seine Adsorptionsfähigkeit für gewisse Farbstoffe. J. König, J. Hansenbaumer und C. Hassler¹⁾ haben Methylviolet, L. Vigonon Fuchsin S. und Pikrinsäure²⁾, F. Schneider Methylenblau B.³⁾ als Farbstoff gewählt. Der Verfasser hat beobachtet, dass für Proben von saurem Boden sich am besten Eosin ($C_{20}H_6O_5Br_4K_2$) eignet, weil dieser Farbstoff seine Färbung in einem schwachen Säure- und Alkalimedium nur schwer verändert.

Die Konzentration des Farbstoffes muss dabei so gewählt werden, dass einerseits nicht aller Farbstoff, anderseits aber doch so viel davon adsorbiert wird, dass ein genügender Unterschied in der Färbung vor und nach der Adsorption deutlich sichtbar ist. Die Bestimmung der Farbstoffkonzentration kann zweckmässig auf kolorimetrischem Wege durch Vergleich mit den Kontroll-lösungen ausgeführt werden. Für die Farbstofflösungen werden drei Konzentrationen, nämlich 0,5, 1,0 und 2,0 g im Liter verwendet. Wenn auch die Temperatur und die Berührungszeit bei der Adsorption keine so grosse Rolle spielen wie bei anderen physikalischen und chemischen Vorgängen, so werden wir doch bei konstanter Temperatur arbeiten und die Farbstoffe stets zu gleicher Zeit einwirken lassen. Das Gemisch von 5 g Boden und 20 ccm Farbstofflösung wird jedesmal ca. 1/2 Stunde geschüttelt, 6 Stunden stehen gelassen und bei Zimmertemperatur (15—17°) geprüft.

Tabelle 8.

Nummer der Proben	Der unadsorbiert gebliebene Farbstoffmenge entsprechend, jede Farbstofflösung in 5 ccm Wasser zurücktitriert. Die Werte bezeichnen die Masse der Farbstofflösungen.			
	geglüht	original		
	1/2‰ Frbst. Lsg.	1/2‰ Frbst. Lsg.	1‰ Frbst. Lsg.	2‰ Frbst. Lsg.
1	0	4,2	5,5	10,0

1) Centrallbl. Agr. Chem., Bielemann, (1912), 217.

2) Ibid, (1911), 649.

3) Internationale Mittl. Bodenk., (1912), 81.

2	1,0	4,5	5,8	9,5
3	1,0	1,5	2,8	2,0
4	—	—	—	—
5	0,5	2,8	2,5	2,5
6	1,0	2,8	2,4	5,0
7	0,5	1,7	1,2	1,5
8	1,0	0,5	1,0	1,0
9	6,0	2,0	2,2	6,8
10	6,0	0,7	0,9	1,1
11	1,0	1,2	2,0	1,7
12	1,0	2,2	3,0	4,7

Für die Herstellung des Adsorptionsgleichgewichts kommen wir dann empirisch zu der Gleichung:

$$-\frac{X}{M} = K C^{\frac{1}{n}}; \quad \frac{X}{M} \text{ (Adsorbiert)} = K C^{1/n} \text{ (frei)}$$

In welcher Gleichung X die Farbstoffmenge, die von 1 mg Boden aus einer Lösung adsorbiert wird und C die Konzentration (mg in 1 ccm) des Farbstoffes in Wasser nach der Adsorption und $\frac{1}{n}$ der Exponent (Vergl. Versuch 5) ist. Für die rechnerische Darstellung der Adsorptionsvorgänge wird obige Gleichung logarithmiert: so erhalten wir

$$\log K = \log \frac{X}{M} - \frac{1}{n} \log C$$

Die folgenden Werte wurden gefunden:

Tabelle 9.

Nummer der Proben	1/2 ‰ Farbstofflösung		1 ‰ Farbstofflösung		2 ‰ Farbstofflösung	
	X/M	C	X/M	C	X/M	C
1	4,08/5000	3,42	7,15/5000	7,85	10,00/5000	20,00
2	3,95/5000	3,55	6,95/5000	8,05	10,34/5000	19,66
3	5,77/5000	1,73	9,65/5000	5,36	21,43/5000	8,57
5	4,81/5000	2,69	10,00/5000	5,00	20,00/5000	10,00
6	4,81/5000	2,69	10,14/5000	4,86	15,00/5000	15,00

7	5,60/5000	1,90	12,10/5000	2,90	23,08/5000	6,92
8	6,89/5000	0,61	12,50/5000	2,50	25,00/5000	5,00
9	5,38/5000	2,14	10,42/5000	4,58	12,50/5000	17,29
10	6,58/5000	0,90	12,71/5000	2,29	24,59/5000	5,41
11	6,05/5000	1,45	10,72/5000	4,28	22,39/5000	7,61
12	5,21/5000	2,29	9,38/5000	5,62	15,46/5000	14,54

Zur Erklärung der Beziehungen, die zwischen den kolloidalen Substanzen, welche aus dem Boden mit Wasser ausgelaugt wurden und der Adsorptionsfähigkeit des Bodens bestehen, wurde folgender Versuch durchgeführt. Als Proben wurden die dialysierten Reste bei dem Versuch gebraucht und nach der oben erwähnten Methode untersucht.

Tabelle 10.

Nummer der Proben	Der unadsorbiert gebliebene Farbstoffmenge entsprechend jede Farbstofflösung in 5 ccm Wasser zurücktitriert Die Werte bezeichnen die Masse der Farbstofflösungen.		
	geglüht	original	
	1/2 960 Farbstofflösung	1/2 960 Farbstofflösung	1 960 Farbstofflösung
1	0	5,0	5,4
2	1,0	5,1	5,9
3	1,0	1,7	2,0
5	0,5	3,5	4,0
6	1,0	4,0	4,5
7	0,5	1,6	1,5
8	1,0	0,9	1,5
9	6,0	2,0	2,5
10	6,0	1,5	1,0
11	1,0	2,0	1,9
12	1,0	2,5	2,9

Die Bestimmung der Werte von X/M und C wurde nach den oben erwähnten Formeln vorgenommen, sie ergab folgende Zahlen :

Tabelle 11.

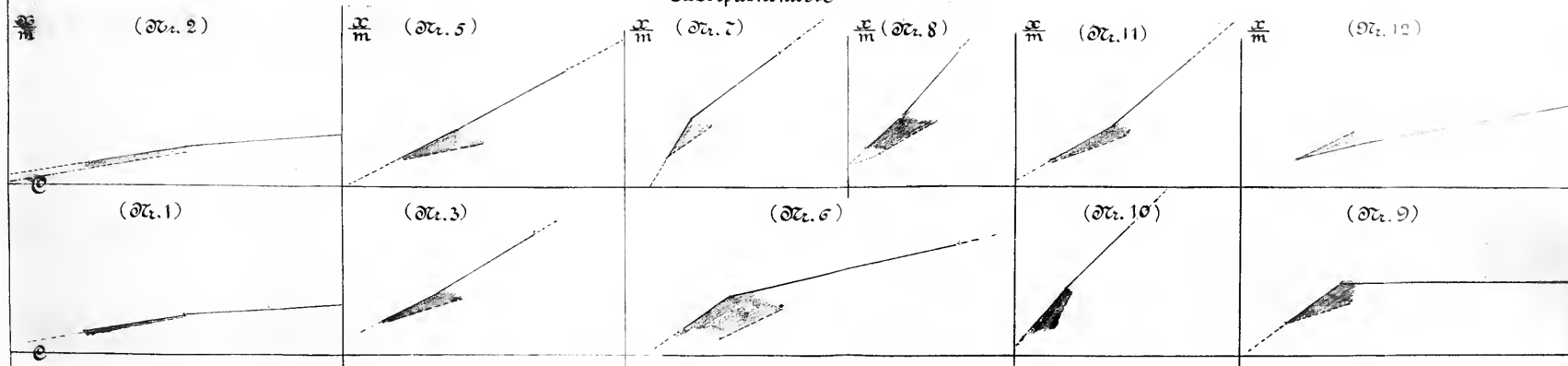
Nummer der Proben	1/2 % Farbstofflösung		1 % Farbstofflösung	
	X/M	C	X/M	C
1	3,72/5000	3,78	7,20/5000	7,80
2	3,83/5000	3,67	6,90/5000	8,10
3	5,60/5000	1,90	10,72/5000	4,28
5	4,50/5000	3,00	8,40/5000	6,60
6	3,30/5000	4,20	7,95/5000	7,05
7	5,70/5000	1,80	11,55/5000	3,45
8	6,37/5000	1,13	11,55/5000	3,45
9	5,40/5000	2,10	10,05/5000	5,00
10	5,77/5000	1,73	12,50/5000	2,50
11	5,40/5000	2,10	10,60/5000	4,40
12	5,03/5000	2,47	9,60/5000	5,40

Wir wollen die graphische Darstellung dieser Vorgänge wählen; dabei ergeben sich die folgenden Daten, wobei in dem rechtwinkligen Koordinatensystem C auf die Abszisse und X/M auf die Ordinate zu stehen kommen.

In der obigen Figur zeigt die Linie (—) die Adsorptionskurve des Originals und die punktierte Linie (-----) die von der Bodenlösung befreiten Bodenreste. Der Raum zwischen den beiden Linien zeigt das Adsorptionsvermögen der kolloiden Substanzen in der Bodenlösung. Dieser Raum ist bei den humusreichen Proben grösser als bei den humusarmen.

Im Einklang mit J. M. B e m m e l n s Angabe nimmt die Adsorptionskraft für Farbstoffe wie Nährlösungen mit der Verdünnung der Konzentration ab. (Vergl. Die Adsorption von W o. O s t w a l d. Dresden 1910)

Absorptionskurve



Vergleichung von Quellungsvolumen und Hygroskopizität mit des Adsorption von Farbstoff.

Tabelle 12.

Proben	X/M•10	Quellungs- volumen.	Hygro- skopizität.	Verhältnisse.	
				(X/M•10) : (Q. V.)	(X/M•10.) : (Hy.)
1	0,81	5,7	3,40	0,142	0,288
2	0,79	5,0	3,26	0,158	0,244
3	1,15	8,0	7,13	0,143	0,216
5	0,96	6,3	4,48	0,152	0,214
6	0,96	6,1	3,54	0,155	0,271
7	1,12	7,7	7,95	0,145	0,141
8	1,37	8,6	10,02	0,159	0,136
10	1,31	7,5	—	0,174	—
11	1,21	6,5	5,42	0,186	0,223
12	1,04	5,7	3,82	0,182	0,272

Die Verhältnisse zwischen Adsorption und Quellungsvolumen kommen einander in allen Proben sehr nahe; sowie auch die zwischen Adsorption und Hygroskopizität (ausser Proben 7 u. 8. (l. c.))

Versuch 5. Ammoniakabsorption.

Über die Absorption von Nährlösung ergeben die Versuche Königs und seiner Mitarbeiter dass die Absorptionskraft, wie bereits J. M. van Bemmeln (l. c.) angibt, mit der Verdünnung der Nährlösung abnimmt. Das Kali wird in lockerer Bindung hauptsächlich vom kolloiden Ton gebunden. Verschiedene Arbeiten behandeln die Absorptionsgleichgewichte für Böden. Nach J. H. Aberson¹⁾ stellt sich bei der Bodenabsorption, welche keine chemische Wirkung im Sinne des Guldberg-Waagéschen Massenwirkungsgesetzes ist, ein Gleichgewicht zwischen absorbierten und ausgetriebenen Ionen ein. Diese Absorption wird von der Oberfläche der Teile im kolloiden Zustand veranlasst und folgt denselben Gesetzen wie

1) Koll. Zeitschr., 10, (1912), 13.

Kohle, Wolle, Seide u. s. w. Mehrere Autoren wie R. Gans¹⁾, K. Glinka²⁾, F. Cornu³⁾ machten Versuche mit natürlichem und künstlichem Zeolith. In jedem Falle ist wie bereits E. Blank⁴⁾ bemerkt, wohl anzunehmen, dass die im Boden auftretenden Absorptionsphänomene an Gele oder an kolloide Substanzen gebunden sind.

Über die Festbindung des Ammoniakstickstoffes hat bereits Th. Pfeiffer⁵⁾ berichtet, dass die absorbierten Ammoniakmengen wenigstens zum Teil nach der Aufnahme durch die Pflanzenwurzel zugänglich bleiben. Andere Autoren wie D. J. Hissink⁶⁾ und R. Gans⁷⁾ haben diese Frage nach verschiedenen Richtungen hin diskutiert. Aber noch ist kein Bericht über die Ammoniakabsorption des sauren Bodens erschienen.

Daher wollen wir folgende Versuchsergebnisse berichten.

Folgende Werte sind von B. Ohashi, welcher unter Anleitung des verstorbenen Prof. Dr. Suzuki gearbeitet hat, festgestellt worden.

Zum Zwecke der Bestimmung der Absorption verfahren wir in folgender Weise: 100 g Erde wurden mit 250 ccm normaler Ammonchloridlösung gemischt, geschüttelt und fünf Tage lang stehen gelassen. Dann wurden 25 ccm verdünnten Extrakts in einen 250 ccm haltigen Messkolben gefüllt. Das Ammoniak wurde bestimmt durch Destillation von 50 ccm Wasser und frisch gebrannter Magnesia. Das Ammoniak wurde mit 1/10 Normalschwefelsäure aufgefangen und mit 1/10 Normalnatronlauge zurücktitriert.

Tabelle 13.

Nummer der Proben	Absorptionskoeffizient.		
	in N/100 NH ₄ Cl Lösungen.	in N/10 NH ₄ Cl Lösungen.	im Normal NH ₄ Cl Lösungen
1	17,55	84,55	183,2
2	16,38	65,99	120,9
3	20,47	111,4	365,3

1) Jahrb. Kgl. Dr. Geol. Landesanst. u. Bergakad., **26**, (1905), 178; **27**, (1906), 63.

2) St. Petersburg., **141**, (1906).

3) Koll. Zeitschr., **5**, (1909), 281; **4**, (1909), 291.

4) Mitt. Landw. Inst. Univ., Breslau, **4**, (1907), 1/2.

5) Ibid., **3**, (1905), 299.

4	14,62	67,0	132,7
5	15,86	64,7	161,2
6	14,62	59,1	156,4
7	17,55	88,9	154,8
8	11,69	46,2	90,5
9	15,86	82,4	227,1
10	—	165,6	428,90
11	16,38	104,7	279,0
12	15,86	75,7	173,9

Für die Herstellung des Absorptionsgleichgewichts erhalten wir folgende Zahlen: (Vergl. Versuch 4).

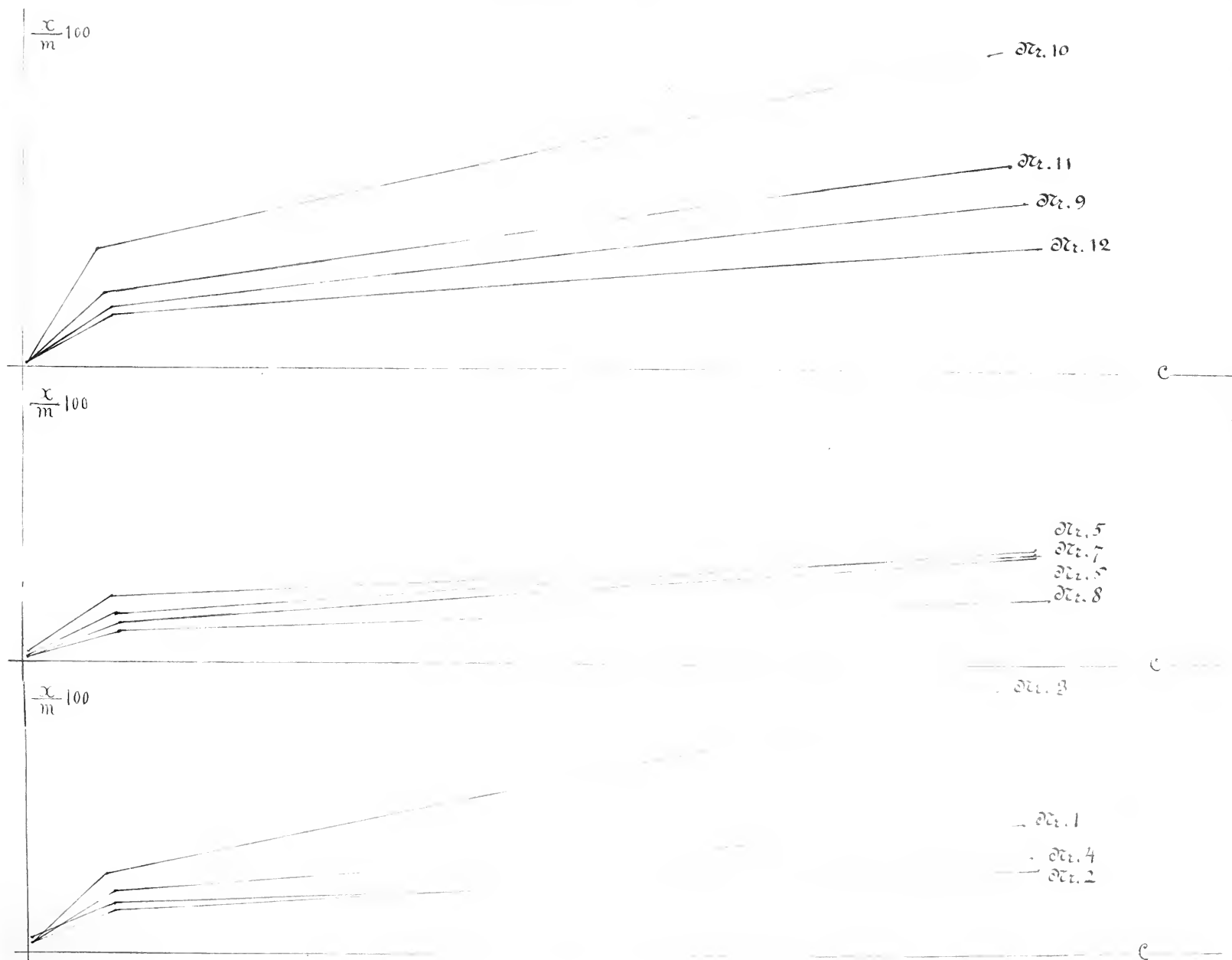
Tabelle 14.

Nummer der Proben	(I) N/100 NH ₄ Cl-Lösung		(II) N/10 NH ₄ Cl-Lösung		(III) Normal NH ₄ Cl-Lösung	
	N/M	C	N/M	C	N/M	C
1	0,000175	0,047	0,000845	1,213	0,081832	13,78
2	0,000164	0,051	0,000659	1,260	0,001209	13,93
3	0,090205	0,035	0,001114	1,146	0,003653	13,34
4	0,000146	0,058	0,000670	1,264	0,001327	13,90
5	0,000158	0,055	0,000647	1,264	0,001613	13,83
6	0,000146	0,058	0,000591	1,302	0,001564	13,84
7	0,000176	0,049	0,000889	1,203	0,001548	13,85
8	0,000117	0,062	0,000462	1,309	0,000905	14,01
9	0,090158	0,055	0,000824	1,219	0,002271	13,65
10	—	—	0,001658	1,011	0,004289	13,16
11	0,000164	0,051	0,001047	1,163	0,002790	13,54
12	0,000158	0,055	0,000757	1,236	0,001739	13,80

Wir wollen die graphische Darstellung dieses Versuches wählen, dabei ergeben sich die folgenden Daten.



Absorptionskurve





Wie die obigen Figuren zeigen, nimmt die Nährstoffabsorptionskraft wie die der Farbstofflösung mit der Verdünnung der Konzentration ab; der Beziehungswert ist in allen Proben gleich.

Aus obigen Zahlen werden die Werte von $\frac{1}{n}$ und K nach folgenden Gleichungen berechnet. Die schon erwähnte Gleichung ist:

$$K = \frac{X/M}{C^{1/n}} \quad ; \quad \frac{X}{M} = K C^{1/n}$$

zuerst wird der Wert von $1/n$ nach folgenden Gleichungen berechnet.

$$\frac{X_1}{M} = K \cdot C_1^{1/n} \quad ; \quad \frac{X_2}{M} = K \cdot C_2^{1/n} \quad ; \quad \frac{X_1}{X_2} = \frac{C_1^{1/n}}{C_2^{1/n}}$$

$$\log X_1 - \log X_2 = \frac{1}{n} (\log C_1 - \log C_2); \quad n = \frac{\log C_1 - \log C_2}{\log X_1 - \log X_2}$$

Tabelle 15.

Nummer der Proben	1/n eliminiert aus (I) u. (II)	1/n eliminiert aus (II) u. (III)	1/n Durchschnittszahl
1	0,1866	0,1908	0,1887
2	0,1628	0,1659	0,1644
3	0,1726	0,2818	0,2272
4	0,2110	0,1800	0,1955
5	0,1781	0,2270	0,2025
6	0,1801	0,2296	0,2049
7	0,2064	0,1513	0,1789
8	0,1876	0,1873	0,1875
9	0,2339	0,2459	0,2399
10	—	0,1940	0,1940
11	0,2737	0,2283	0,2510
12	0,2124	0,2057	0,2091
Durchschnittszahl von 1/n der 12 Proben ——— 0,2036			

Dann folgt die Berechnung des Wertes von K mit obiger Durchschnittszahl nach folgenden Gleichungen.

$$K = \frac{X/M}{C^{1/n}} \quad , \quad \log K = \log X/M - 1/n \log C$$

Tabelle 16.

Nummer der Proben	„K“ eliminiert aus (I)	„K“ eliminiert aus (II)	„K“ eliminiert aus (III)
1	0,0003565	0,0008146	0,001117
2	0,0002691	0,0006385	0,000784
3	0,0005882	0,0001082	0,002105
4	0,0002715	0,0006493	0,000796
5	0,0002843	0,0006175	0,000949
6	0,0003842	0,0005593	0,000717
7	0,0003982	0,0008608	0,000979
8	0,0003408	0,0004393	0,000555
9	0,0002007	0,0007754	0,001212
10	—	0,001638	0,002568
11	0,0003466	0,001001	0,001458
12	0,0004145	0,0007242	0,001005
Durch- schnittszahl	0,0003504	0,0008165	0,001038

Aus diesen Ammoniakabsorptionsversuchen lässt sich folgende Zusammenfassung gewinnen.

- 1) Die Ammoniakabsorptionskraft hat keine regelmässige Beziehung zur Farbstoffadsorption. (Vergl. Tabelle 7 mit den Tabellen 13 und 14.)
- 2) Die Ammoniakabsorptionskraft hat keine regelmässige Beziehung zum Quellungsvolumen und der Hygroskopizität. (Vergl. Tabelle 7 mit der Tabelle 13 und 14.)
- 3) Die Ammoniakabsorptionskraft steht nicht in so naher Beziehung wie die Farbstoffadsorptionskraft zu der Oberfläche der Teile im kolloiden

Zustande. Diese Erscheinungen entsprechen wahrscheinlich dem Unterschied der Ammoniak- und Farbstoffadsorptionsphänomene der Gele oder kolloiden Substanzen.

4) Die Konstante des Adsorptionsgleichgewichtes variiert infolge der Konzentrationsveränderungen der Ammoniaksalze, wie die Durchschnittszahlen 0,0003504 in N/100 Lösungen, 0,0008165 in N/10 Lösungen und 0,001038 in Normallösungen zeigen.

5) Die Konstante des Adsorptionsgleichgewichtes des humusarmen sauren Bodens ist grösser als die des humusreichen sauren Bodens. (l. c.)



Zur Erkennung des Unterschiedes der Ammoniakabsorptionsfähigkeit des Bodens nach gebundenen Anionen dient die Vergleichung der Ammoniakabsorptionskoeffizienten von Normalammonchlorid und Normalammonphosphatlösungen.

Tabelle 17.

Nummer der Proben	Ammoniakabsorptionskoeffizient		Unterschied.
	im $\text{NH}_4 \text{Cl}$ Lösungen	im Ammonphosphat-Lsg.	
1	183,2	239,9	56,7
2	120,9	237,3	116,4
3	365,3	352,9	-(13,7)
4	132,7	284,3	141,6
5	161,2	279,0	117,8
6	156,4	239,8	83,4
7	154,8	476,8	322,0
8	90,5	304,4	213,9
9	227,1	472,5	245,4
10	428,9	681,7	252,8
11	219,6	416,5	197,3
12	173,9	303,0	129,1

Aus der Tabelle erkennen wir, dass die Ammoniakabsorptionskoeffizienten der Böden in Normalammonphosphatlösungen grösser sind als in Normalammonchloridlösungen. Der Unterschied zwischen beiden Resultaten beträgt 31 -- 208 %, durchschnittlich 100 %. Zur Diskussion dieser Erscheinungen vergleichen wir die vergrösserten Werte mit dem Aluminiumgehalt des Ammonchloridextrakts des Bodens, wobei sich die folgenden Daten ergeben. Das Aluminium wurde durch Vermischung mit überschüssigem Ammoniak gefällt. Die folgenden Zahlen geben den Gehalt für 100 g Boden.

Tabelle 18.

Nummer der Proben	Unterschied. (I)	Aluminiumgehalt in NH_4Cl -Extrakt von 100 Gramm Erde. m. g. (II)	Verhältnisse (I)/(II)
1	56,7	26,0	2,1
2	116,4	48,4	2,0
3	-(13,7)	(162,0)	?
4	141,6	75,5	2,0
5	117,8	67,0	2,0
6	83,4	59,5	1,5
7	322,0	59,4	(5,4)
8	213,9	59,3	3,6
9	245,4	82,0	2,9
10	252,8	101,1	2,5
11	197,3	102,1	2,0
12	129,1	93,7	1,4

Die Tabelle zeigt, dass die vergrösserten Werte der Ammoniakabsorptionskoeffizienten aller Proben (ausser 3 und 7,) wahrscheinlich in nahem Verhältnis stehen, und es ist wohl anzunehmen, dass die nach folgende Tatsache wenigstens als ein wichtiger Faktor für die Absorptionskraft erscheint. Bei Ammonphosphatbehandlung ist die befreiende Phosphorsäure mit Aluminium verbunden und diese unlöslichen Verbindungen vergrössern

die Ammoniakabsorptionskraft des Bodens.

Zusammenfassung.

1) Die Resultate der Quellungsversuche zeigen uns, dass die gebildeten Gelformen je nach der Art des Reagens verschieden sind und die Gelbildungsmaterialien des humusarmen sauren Bodens nicht einheitliche Substanzen darstellen.

2) Die Unterschiede der vergrößerten Volumen zwischen den originalen Proben und den mit Wasser extrahierten Resten sind sehr klein im humusarmen Boden. Daher haben die kolloiden Substanzen, die aus dem Boden mit reinem Wasser extrahiert werden, keine wichtige Wirkung auf die Quellung des Bodens.

3) Die Quellungswärme ist wahrscheinlich proportional dem Quellungsgrade für humusarmen Boden.

4) Die Hygroskopizität steht gewöhnlich in nahen Beziehungen zu dem Quellungsgrade.

5) Im Einklang mit J. M. van Bemmels Angabe nimmt die Adsorptionskraft für Farbstoffe wie auch Nährlösungen mit der Verdünnung ihrer Konzentration ab. Die Verhältnisse zwischen Farbstoffadsorption und Quellungsvolumen kommen sich sehr nahe wie auch die zwischen Farbstoffadsorption und Hygroskopizität.

6) a. Die Ammoniakabsorptionskraft hat keine regelmässige Beziehung zu den Farbstoffadsorptionen.

b. Die Ammoniakabsorptionskraft hat keine regelmässige Beziehung zum Quellungsvolumen und zu der Hygroskopizität.

c. Die Ammoniakabsorptionskraft steht nicht in so naher Beziehung wie die Farbstoffadsorptionskraft zu der Oberfläche der Teile im kolloiden Zustande. Diese Erscheinungen beziehen sich wahrscheinlich auf die Gele oder kolloiden Substanzen.

d. Die Konstante des Absorptionsgleichgewichtes variiert infolge der Konzentrationsveränderungen der Ammoniaksalze, wie die Durchschnittszahlen 0.0003504 in N/100 Lösungen, 0,0008165 in N/10 Lösungen

und 0,0001038 in Normallösungen zeigen.

e. Die Konstante des Absorptionsgleichgewichtes des humusarmen sauren Bodens ist grösser als die des humusreichen Bodens.

7) Der Ammoniakabsorptionskoeffizient der Böden in Normalammonphosphatlösungen ist grösser als der für Normalammonchloridlösungen. Und es ist wohl anzunehmen, dass bei Ammonphosphatbehandlung die freigewordene Phosphorsäure sich mit dem Aluminium verbindet, und diese unlösliche Verbindung die Ammoniakabsorptionskraft des Bodens vergrössert.

(H. Z. D.)

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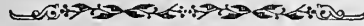
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ÜBER DIE CHRYSOPIDEN-FAUNA JAPANS

von

H. Okamoto, *Nōgakushi*

ENTOMOLOGEN AN DER HOKKAIDO LANDW. VERSUCHSSTATION

(Mit I Figur im Text)

Die Neuropteren in Japan wurden schon in der ersten Hälfte des vorigen Jahrhunderts von C. P. Thunberg, H. Burmeister und M. P. Rambur und seitdem von zahlreichen andern Entomologen weiter studiert. Da sie aber immer nur teilweise untersucht wurden, fehlt es an einer umfassenden Kenntnis der ganzen Ordnung, besonders die Chrysopiden sind noch nicht eingehend studiert worden.

Die Chrysopiden Japans sind bis jetzt von den Entomologen H. Burmeister,¹⁾ Fr. Warker,²⁾ R. Mac Lachlan,³⁾ A. Gerstaecker,⁴⁾ L. Navás,⁵⁾ S. Matsumura,⁶⁾ H. Okamoto,⁷⁾ und E. Petersen,⁸⁾

-
- 1) Handbuch der Entomologie, 2. Bd. 1839.
 - 2) List of the Specimens of Neuropterous Insects in the Collection of the British Museum, part II. (Sialidae, Nemopterides). 1853.
 - 3) a, New Genera and Species, &c., of Neuropterous Insects (Linn. Proc. Zoology, Vol. IX. 1867).
b, A Sketch of our present Knowledge of the Neuropterous Fauna of Japan (Trans. Ent. Soc., London, Pt. II. 1875).
 - c, Trans. Ent. Soc., London, 1893, P. 230).
 - 4) Über neue und weniger bekannte Neuropteren aus der Fam. Megaloptera Burm. (Mitt. naturw. Vereins f. Neu-Vorpommern u. Rügen. 1893).
 - 5) a, Crisópidos Nuevos (Brotéria Serie Zool., Vol. IX, Fasc. I. 1910).
b, Quelques Névroptères de la Sibérie méridionale-orientale (Revue Russe d'Entom., XII. 1912).
 - 6) Erster Beitrag zur Insekten-Fauna von Sachalin (Jour. Coll. Agr. Tohoku Imp. Univ., Sapporo, Vol. IV, Pt. I. 1911).
 - 7) Eine neue Chrysopiden-Art Japans (Trans. Sapporo Nat. Hist. Soc., Vol. IV, Pt. I. 1912).
 - 8) H. Sauters Formosa-Ausbeute.....Planipennia II, Megaloptera and Mecoptera (Ent. Mitt., Bd. II, No 9. 1913).

[Jour. of the College of Agr., Tohoku Imp. Univ., Sapporo, Vol. VI. Pt. 3, August 1914.]

behandelt worden; im ganzen sind etwa 20 Arten näher bekannt. Hiermit ist jedoch die Zahl der in Japan einheimischen Arten bei weitem noch nicht erschöpft.

Aus meinen bisherigen Untersuchungen geht hervor, dass in Japan nicht weniger als 30 Arten vorkommen, von denen 11 neu sind. Sie verteilen sich auf 5 Gattungen, darunter ist eine Gattung (*Pseudochrysa*) neu.

Japan liegt zwischen 22 und 50° N. Br. Das Land ist sehr lang und schmal. Es besteht aus 6 grossen Inseln (Sachalin, Hokkaido, Hondo, Shikoku, Kiushiu, Formosa), einer grossen Anzahl kleiner Inseln und einer Halbinsel (Korea). Deshalb muss das Studium der japanischen Fauna wissenschaftlich sehr interessant sein. Das Material, welches ich in dieser Arbeit erwähne, gehört grösstenteils der palaearktischen Fauna an. Die Insel Formosa, als Grenzgebiet der orientalischen Region, besitzt eine Fauna, welche sowohl die zur orientalischen, als auch die zur palaearktischen Region gehörigen Formen umschliesst. Diese Tatsache ist von einem grossen biographischen Interesse.

Herr Prof. Dr. S. M a t s u m u r a an der landwirtschaftlichen Fakultät der Kais. Tohoku Universität zu Sapporo hat mir sein reiches Material an Chrysopiden zum Studium überlassen und mir dabei mit freundlichem Rat beigestanden, wofür er meinen herzlichsten Dank entgegennehmen möge.

Tabelle zur Bestimmung der Gattungen.

- | | | |
|----|---|-------------------------|
| 1. | Vorderflügel mit der 3. Kubitalzelle | 2. |
| | Derselbe ohne solche | <i>Nacaura</i> Nav. |
| 2. | Die 3. Kubitalzelle oval | 3. |
| | Die 3. Kubitalzelle viereckig | <i>Nothochrysa</i> M'L. |
| 3. | Kosta am Grunde im Bogen nach vorn abbiegend | <i>Ancylopteryx</i> Br. |
| | Kosta am Grunde erst später abbiegend | 4. |

- 4, Abdomen des Männchens an der Spitze mit einem paarigen, zangenförmigen Anhang *Pseudochrysa* nov. gen.
 Abdomen des Männchens ohne solchen *Chrysopa* Lch.

I. Gatt. **Nakaura** Nav.

Navás, Revue Russe d'Entom., 1913, XIII, No. 2, p. 280.

In Japan kommt nur eine Art vor.

1. **Nakaura Matsumurae** Okam.

Apochrysa Matsumurae Okamoto, Trans. Sapporo Nat. Hist. Soc., Vol. IV, 1912, p. 13, Fig. 1.

Hab.....Kiushiu (Kagoshima).

Diese Art kommt in Japan wahrscheinlich sehr selten vor.

II. Gatt. **Nothochrysa** M'L.

Mac Lachlan, Trans. Ent. Soc., London, 1868, p. 195.

In Japan kommen zwei Arten vor.

Uebersicht der Arten.

Pronotum an den Seiten rot gefleckt *japonica* M'L.

Pronotum einfarbig grün *olivacea* Gerst.

1. **Nothochrysa japonica** M'L.

Mac Lachlan, Trans. Ent. Soc., 1875, p. 182.

Hab.....Hondo (Kyoto, Gifu, Prov. Kii), Shikoku (Prov. Iyo), Kiushiu (Kumamoto, Nagasaki) und Formosa (Shinsha, Kankau, Taihanroku, Arisan).

Diese ist eine schöne und weit verbreitete Art und kommt in Süd-Japan und Formosa vor. In Nordost-Japan und auch in Hokkaido ist sie noch nicht bekannt. Es könnte m. E. Zentral-Hondo als die Nordgrenze ihrer Verbreitung bezeichnet werden. Obwohl diese Art sich in der palaearktischen Region befindet, gehört sie eigentlich doch zur orientalischen.

2. **Nothochrysa olivacea** Gerst.

Gerstaecker, Mitt. Neu-Vorpom. u. Rügen, 1893, p. 74.

Hab.....Hondo.

Nach Gerstaecker wurde diese Art in Yokohama gefunden. Aber es ist sehr zweifelhaft, ob sie tatsächlich nach seiner Beschreibung zur Gattung *Nothochrysa* gehört. Da ich leider keine Gelegenheit gehabt habe, dieselbe zu sehen, habe ich sie in die Gattung *Nothochrysa* eingeordnet. Diese Art kommt wahrscheinlich sehr selten in Japan vor.

III. Gatt. **Ancylopteryx** Br.

Brauer, Wien Z. B. Ges. XIV, 1864, p. 901.

Diese Gattung stammt aus Afrika, Südasien und Neufundland; in Japan (Formosa) kommt nur eine Art vor.

1. **Ancylopteryx octopunctata** F.

Hemicrobius octopunctata Fabricius, Ent. Syst. II, 1893, p. 85.

Hab.....Formosa (Kosempo, Sokutu, Taihorin, Hokuto, Ako, Horisha).

Sonstige Fundorte. Cochinchina, Philippinen, Java, Ceylon, Indien, u. s. w.

Diese schöne Art ist nur in der orientalischen Region verbreitet.

IV. Gatt. **Pseudochrysa** nov. gen.

Typus: *Pseudochrysa formosana* nov. gen. et nov. spec.

Oberlippe ganz. Fühler lang und dünn, das erste Basalglied sehr gross. Zwischen den Fühler-Insertionen kein Horn. Flügel breit, mit fast abgerundeter Spitze. Kosta am Grunde gerade, erst später abbiegend, Kostalfeld am Grunde sich verengend. Die dritte Kubitalzelle wird durch den *Ramus divisorius* in zwei ungleiche Hälften geteilt, von denen die obere oval und viel kleiner als die untere ist. Zwei Reihen von *Venulae gradatae* in beiden Flügeln. Abdomen des Männchens an der Spitze mit einem paarigen, schlanken, zangenförmigen Anhang. Klauen an der

Basis breiter werdend.

Diese Gattung steht der Gattung *Chrysopa* sehr nahe.

In Japan kommt nur eine Art vor.

1. ***Pseudochrysa formosana*** nov. spec.

Grün. Gesicht hellgelbbraun, Stirn an den vorderen Seiten je einen purpurroten querstreifartigen Fleck zeigend, Wangen mit je einem schwarzen Fleck. Zwischen den Augen und Fühler-Insertionen purpurrot. Beide Taster hellbraungelb. Fühler viel länger als der Vorderflügel, hellgelbbraun, das erste grosse Basalglied äusserlich purpurrot gefleckt. Pronotum viel breiter als lang, fast viereckig; gelbbraun, fein behaart. *Scapulae anteriores* des Mesothorax schwarz. Beine hellgrün, Schienen am äussersten Ende samt den Tarsen hellgelbbraun, Klauen braun; an der Basis breiter werdend.

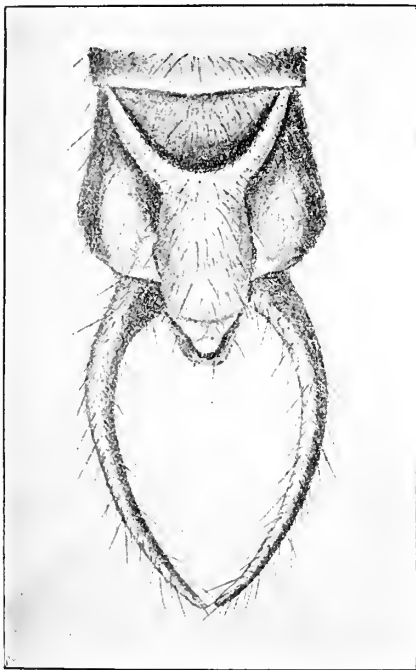


Fig. 1.
Pseudochrysa formosana n. sp.
Hinterleibsende von unten gesehen.

Flügel mit fast abgerundeter Spitze, glashell, Pterostigma fast farblos. Vorderflügel an der Basis purpurrot gefleckt. Randmal und Adern ganz grün, äussere *Venulae gradatae* schwärzlichbraun. *Venulae gradatae* 6/6 (vordere/hintere Reihe) bis 5/5 im Vorderflügel; 3/3 bis 4/4 im Hinterflügel. Kostalqueradern der Vorderflügel bis zum Pterostigma 19 bis 21; zwischen Radius und Radialsektor 9 bis 10 Queradern. Abdominalspitze beim Männchen mit einem paarigen, schlangen, lang behaarten, zangenförmigen Anhang.

Körperlänge	♂ 7 (mit Ausnahme des Anhangs)	♀ 7 mm.
Vorderflügelänge	9	11 mm.
Hinterflügelänge	8	10 mm.
Hab. Formosa (Ako, Taihoku und Shoka), 7 Exemplare, von Prof. S. Matsumura gesammelt.		

V. Gatt. **Chrysopa** Lch.

Leach, Encycl. IX, 1815, p. 138.

In Japan kommen 26 Arten vor, 10 Arten davon sind neu.

Übersicht der Arten.

- | | |
|--|---------------------------|
| 1. Das 2. Basalglied der Fühler ganz schwarz gefärbt | 2. |
| Dasselbe nicht ganz gefärbt oder teilweise (hauptsächlich nach aussen) gefärbt | 6. |
| 2. Zwischen den Fühler-Insertionen ein schwarzer x-förmiger Fleck | 3. |
| Ohne solchen zwischen den Fühler-Insertionen..... | 5. |
| 3. Occiput mit vier schwarzen Pünktchen <i>intima</i> Mac Lachl. | |
| Occiput ohne Pünktchen, ganz schwarz | 4. |
| 4. Mesothorax oben an den Seiten mit breitem schwarzem Saum .. | |
| <i>nigriceps</i> n. sp. | |
| Mesothorax oben mit zahlreichen Flecken | <i>perla</i> L. |
| 5. Occiput mit vier schwarzen Punkten | <i>lezcyi</i> Nav. |
| Occiput ohne Punkte | <i>sapporensis</i> n. sp. |
| 6. Adern einfarbig, grün bis gelb | 7. |
| Queradern ganz oder teilweise schwarz gefleckt | 17. |
| 7. Kopf schwarz oder rot gefleckt | 8. |
| Kopf nicht gefleckt | 11. |
| 8. Kopf zwischen den Fühlern mit einem schwarzen, fast x-förmigen Fleck | <i>furcifera</i> n. sp. |
| Kopf zwischen den Fühlern ohne solchen | 9. |
| 9. Körper mit gelber Längslinie | 10. |

- Körper ohne solche *anpingensis* Peters.
10. Stirn und Clypeus an den Seiten mit je einem schwarzen Streifen
..... *bouincensis* n. sp.
Stirn und Clypeus ohne solchen *vulgaris* var. *microcephala* Br.
11. Körper länger als 10 mm 12.
Körper kürzer als 10 mm 15.
12. Maxillartaster schwärzlichbraun *iuoruata* Mats.
Maxillartaster gelb 13.
13. Fühler etwas länger als Vorderflügel ... 14.
Fühler viel kürzer als Vorderflügel ... *vittata* Wesm.
14. Pronotum länger als breit *ogasarawensis* n. sp.
Pronotum breiter als lang *remota* Wk.
15. Fühler länger als Vorderflügellänge *formosaua* Peters.
Fühler kürzer als Vorderflügellänge 16.
16. Kopf gelbbraun *ruficeps* Mac Lachl.
Kopf rostfarbig *basalis* Wk.
17. Kopf und Thorax mit zahlreichen schwarzen Pünktchen 18.
Kopf und Thorax ohne solche, blutfarbig gefleckt *uippoucensis* n. sp.
18. Taster hell, oder teilweise braun gefleckt 19.
Taster schwärzlichbraun bis schwarz 23.
19. Fühler viel kürzer als Vorderflügel 20.
Fühler entweder gleichlang oder länger als Vorderflügel 21.
20. Vor der Fühler-Insertion ein bogenartiger Fleck... *bipunctata* Burm.
Vor der Fühler-Insertion ohne solchen *cognata* Mac Lachl.
21. Mesonotum und *Scapulae anteriores* dunkelbraun 22.
Mesonotum gelb *sauteri* Peters.
22. Queradern meistens braun bis schwärzlichbraun *decorata* Peters.
Queradern meistens hell *matsunurac* n. sp.
23. Pronotum mit zahlreichen schwarzen Pünktchen 24.
Pronotum ohne solche 25.
24. Acht Punkte im Pronotum *sachalinensis* Mats.

- Sechs Punkte im Pronotum *nikkoënsis* n. sp.
 25. Queradern fast ganz schwarz *cognatella* n. sp.
 Queradern (nur *Venulae gradatae*) schwarz *kurisakiana* n. sp.

1. ***Chrysopa intima*** Mac Lachl.

Mac Lachlan, Trans. Entom. Soc., Lond., 1893, p. 230.

Chrysopa perla L. var. *fracta* Nav., Brotéria Serie Zool., Vol. IX, fasc. 1, 1910, p. 39.

Hab. . . . Sachalin, Hokkaido (Sapporo, Berg Moiwa und Maruyama bei Sapporo, Jozankei, Ishiyama, Yubari), Hondo (Yatsugadake-Berg in der Prov. Shinano, Kofu in der Prov. Kai, Towada-See bei Aomori).

Sonstige Fundorte. Ost-Sibirien (nach Navás) und Sibirien (nach Mac Lachlan).

Diese Art kommt nur im nördlichen Teil von Japan vor, besonders in Hokkaido, seltener in den hohen Gebirgsdistrikten der Provinzen Shinano und Kai. Es scheint mir der Berg Yatsugadake, wo M. Shibakawa sie gesammelt hat, vielleicht die Südgrenze ihrer Verbreitung zu sein. Diese Art findet sich nur in der palaearktischen Region. *Chrysopa fracta* Navás ist ein Synonym für diese Art, trotzdem die Beschreibung darüber zu einfach ist, um die erstere zu identifizieren.

2. ***Chrysopa nigriceps*** nov. spec.

Grasgrün. Kopf mit grasgrünen Flecken, schwarz. Scheitelfleck fast langoval, Stirnfleck fast dreieckig, Seitenflecke der Stirn streifenartig. Fühlerinsettionen grasgrün umkreist. Mundteil braun. Beide Taster schwärzlichbraun, jedes Glied am äussersten Ende gelb. Fühler kurzer als der Vorderflügel, blassbraun, nach der Spitze zu dunkler; das 1. grosse Basalglied hellgelbbraun, das 2. schwarz geringelt. Gula und Collum schwarz. Pronotum breiter als lang, nach vorn etwas verjüngt und abgerundet, schwarz und kurz behaart; an den Seiten je zwei grosse schwarze

Flecke, von denen ein vorderer Fleck fast viereckig und beträchtlich kleiner ist als der andere; Mittelquerfurche ziemlich tief. Prosternum schwarz. Meso- und Metathorax oben an den Seiten mit je einem dicken und schwarzen Streifen, Mesonotum und Metaphragma am vordern Teile schwarz gesäumt. Metasternum schwarz. Meso- und Metathorax hinten mit einem schwarzen x-förmigen Fleck. Abdomen hinten, die Spitze ausgenommen, schwarz; kurz und ziemlich dicht schwarz behaart. Beine mit schwarzer, nicht so dichter Behaarung, grasgrün, Schenkel und Schienen je am Apicalende hellbraun, Tarsen hellbraun, Coxen hinten mit je einem fast rundlichen braunen Fleck, Klauen braun.

Flügel mit rundlicher Spitze, glashell, Pterostigma blassgrün. Längsader und Randmal grün. Im Vorderflügel fast alle Queradern (im Pterostigma braun), *Venulae gradatae*, Radialsektor an der Basis und die *Furcula* schwarz. Im Hinterflügel Kostalqueradern (im Pterostigma grün), Queradern zwischen Radius und Radialsektor, zwischen Radialsektor und Media an der Basis, und zwischen Media und Cubitus, sowie auch *Venulae gradatae* schwarz. *Venulae gradatae* 7/9 bis 8/10 im Vorderflügel; 6/8 bis 7/9 im Hinterflügel. Kostalqueradern des Vorderflügels bis zum Pterostigma 27 bis 28; zwischen Radius und Radialsektor 13 bis 14 Queradern.

Körperlänge	10 bis 12 mm.
Vorderflügelänge	14 bis 17 mm.
Hinterflügelänge	13 bis 16 mm.

Hab. Yatsugadake-Berg in der Prov. Shinano, (drei Exemplare am 22. Juli 1911), und Kamikochi in der Prov. Shinano, (zwei Exemplare am 9. Juli 1912), gesammelt von M. Shibakawa.

Diese Species steht *Chrysopa perla* L. und der *C. intima* Mac Lachl. nahe, unterscheidet sich jedoch von den beiden durch den Kopf- und Brustfleck.

3. *Chrysopa perla* (L.).

Hemcrobis perla L., Syst. Nat. Ed. 12, 1. 2, 1768, 911, 2.

Hab..... Hondo (am Ose-See in der Prov. Kōzuke).

Sonstige Fundorte...Europa, Sibirien (nach Mac Lachlan und Navás).

Diese Species ist sehr weit verbreitet in der palaearktischen Region, in Japan aber sehr selten.

4. **Chrysopa lezeyi** Nav.

Navás, Brotéria Ser. Zool., Vol. IX, fasc. 1, 1910, p. 42.

Hab..... Hokkaido (Sapporo), Hondo (Kōfu? nach Navás).

Diese Art wurde von Navás ohne exakte Lokalitätsangabe ihres Vorkommens in Japan bekannt gemacht. Ich besitze nur ein Stück aus Hokkaido. Es ist anzunehmen, dass sie auch in den Gebirgsdistrikten Shinano und Kai von Zentralhondo vorkommt, aber jedenfalls sehr selten. Dieselbe gehört in die palaearktische Region.

5. **Chrysopa sapporensis** nov. spec.

Grün, grünlichgelb bis gelb. Kopf mit neun schwarzen Fleckchen: das eine zwischen den Fühlern gross; die zwei vor dem Fühler etwas gebogen, eins in jeder Wange, zwei ziemlich lange an den Seiten des Clypeus, und zwei am Scheitel. Beide Taster mit gelbbrauner Spitze, schwärzlichbraun. Fühler beträchtlich kürzer als der Vorderflügel, gelbbraun, nach der Spitze zu brauner; das 1. grosse Basalglied wie der Kopf gefärbt, das 2. schwarz geringelt. Pronotum breiter als lang, nach vorn verjüngt und gerundet, mit einer tiefen Querfurche; an den Seiten mit je zwei braunen (zuweilen undeutlichen) Flecken; schwärzlich kurz behaart. *Scapulae anteriores* mit je zwei schwärzlichbraunen Punkten, Metanotum mit einem gleichfarbigen Punkte. Abdomen und Beine schwarz behaart; Beine wie der Körper gefärbt, nur die Schienen am äussersten Ende und die Klauen braun.

Flügel mit rundlicher Spitze, glashell, Pterostigma hellgelbbraun, Längs- und Queradern grün; alle Kostalqueradern in beiden Flügeln je an ihrer

Basis, Queradern zwischen Radius und Radialsektor an ihrem Ursprung, und einige Basalqueradern der Vorderflügel schwarz; Randmal und Adern schwarz, ziemlich dicht behaart. *Venulae gradatae* 8/9 bis 7/8 im Vorderflügel; 7/8 im Hinterflügel. Kostalqueradern des Vorderflügels bis zum Pterostigma 24 bis 26; zwischen Radius und Radialsektor 12 bis 13 Queradern.

Körperlänge	8 bis 9 mm.
Vorderflügelänge	12 bis 13 mm.
Hinterflügelänge	11 bis 11.5 mm.

Hab..... Hokkaido (Sapporo, Muroran, Jozankei, Kosawa), Hondo (Takasago). In Hokkaido von Juni bis September sehr häufig.

Diese Art steht *Chrysopa novempunctata* Navás und *Ch. lezei* Navás sehr nahe, unterscheidet sich jedoch von der ersteren durch den Fleck der Adern und des Thorax, von der letzteren durch den Fleck des Kopfes und des Thorax.

6. *Chrysopa furcifera* Mats. (n. l.), nov. spec.

Grün mit gelber Mittellinie. Kopf zwischen den Fühlern mit einem schwarzen x-förmigen Flecke, gelblich. Wange mit einem schwarzen Streifen. Scheitel zwischen dem Scheitelfleck und Augen rötlich. Maxillartaster schwarz glänzend, Labialtaster schwärzlichbraun. Fühler fast so lang wie die Vorderflügelänge, dunkelbraun, nach der Basis zu heller; das 1. und 2. gelbe Basalglied aussen streifenartig schwarz gefleckt, bei einigen Stücken das grosse 1. innen mit einem langovalen schwärzlichbraunen Flecke. Prothorax breiter als lang, an den Seiten fast parallel und nach vorn zu gerundet; oben an den Seiten sehr breit schwärzlichbraun gesäumt; schwarz, kurz behaart.

Flügel mit zugespitzter Spitze, glashell, Pterostigma lang, grünlich. Randmal, Längs- und Queradern grüngelb; ziemlich dicht und struppig behaart. *Venulae gradatae* 7/9 bis 10/9 im Vorderflügel; 7/8 bis 10/10 im Hinterflügel. Kostalqueradern des Vorderflügels bis zum Pterostigma

23 bis 25; zwischen Radius und Radialsektor 16 bis 17 Queradern. Beide Flügel sehr schmal und lang.

Körperlänge 9 bis 11 mm.

Vorderflügelänge 12 bis 14 mm.

Hinterflügelänge 11 bis 13 mm.

Hab.....Formosa (Shirin), Riukiu (Insel Loo-choo), Kiushiu (Nagasaki), Hondo (Suma, Kumayama). Mai bis Oktober ziemlich häufig. Gesammelt von Prof. S. Matsumura, K. Kuroiwa, Y. Horikawa, M. Shibakawa und Kimura.

Diese Art gehört zur *Vulgarisgruppe* und ist durch den Kopffleck sehr leicht kenntlich.

7. **Chrysopa anpingensis** Peters.

Petersen, Ent. Mitt., II, Nr. 9, 1913, p. 259.

Hab.....Formosa (Anping, Tainan).

Diese Art kommt nur in Formosa vor und zwar nicht häufig.

8. **Chrysopa boninensis** nov. spec.

Diese Art steht *Ch. anpingensis* Petersen (aus Formosa) nahe.

Grün mit einer gelben, ziemlich dicken Mittellinie. Gesicht gelb; Stirn am vorderen Rande mit zwei schwärzlichbraunen, etwas schrägen Querstreifen, Wangen mit einem schwärzlichbraunen Punkte, Clypeus an den Seiten mit je einem schwärzlichbraunen, etwas dicken Streifen. Maxillartaster schwarz, jedes äusserste Glied schwärzlichbraun. Labialtaster gelbbraun. Fühler etwas länger als der Vorderflügel, gelbbraun, nach der Spitze zu dunkler, das 1. und 2. Basalglied grüngelb. Prothorax breiter als lang, fast viereckig, mit einer Quercarina fast in der Mitte und einer tiefen Mittelfurche hinter der Quercarina; schwarz, fein behaart. Beine hellgrün, Tarsen hellbraun; kurz, ziemlich dicht und schwärzlichbraun behaart.

Flügel am Ende zugespitzt, glashell, Pterostigma fast undeutlich, Randmal und Adern grün, struppig, dicht behaart. *Venulae gradatae* 5/7

bis 7/9 im Vorderflügel; 4/7 bis 6/9 im Hinterflügel. Kostalqueradern des Vorderflügels bis zum Pterostigma 18 bis 21; zwischen Radius und Radialsektor 11 bis 13 Queradern.

Körperlänge	8 mm.
Vorderflügelänge	11 bis 12 mm.
Hinterflügelänge	10 bis 11 mm.

Hab. Formosa (Tainan, Ako und Koshun); Insel Bonin, zwei Exemplare am 20. August 1905 von Prof. S. Matsumura, und zwei Exemplare im Juli 1911 von I. Kuwana gesammelt.

9. **Chrysopa vulgaris** Schneider var. **microcephala** Br.

Brauer, Haid. Abhdl. IV. 6. 4.

Hab. Hondo (Kyoto).

Sonstiger Fundort. Europa.

Im Jahre 1875 wurde diese Art von Mac Lachlan identifiziert, er hat sie aber mit Fragezeichen? versehen, da das betreffende Stück nicht vollkommen war.

Nach meinen zahlreichen Exemplaren zu schliessen, gehört diese Art zweifellos zu *Ch. vulgaris* Schn. var. *microcephala* Br., was aber bezüglich der Verbreitungsverhältnisse einige Schwierigkeiten bietet.

10. **Chrysopa inornata** Mats.

Matsumura, Jour. Coll. Agr. Tohoku Imp. Univ., Sapporo, Vol. IV, Pt. I, 1911, p. 14.

Hab. Sachalin und Hondo (Kamikochi in der Prov. Shinano).

Diese Art wurde zuerst als eine in Sachalin heimische veröffentlicht, aber sie kommt auch in Mitteljapan, namentlich Shinano vor, wo M. Shibakawa einige Exemplare gesammelt hat. Zentraljapan muss wohl als die Südgrenze des Verbreitungsgebietes dieser Art bezeichnet werden.

11. **Chrysopa vittata** Schneid.

Schneider, Monogr. Chrys., 1815, p. 65, 1. Pl. 7.

Hab. Hokkaido (Sapporo).

Sonstiger Fundort. Europa.

Ausser in Hokkaido wurde sie in Japan noch nicht gesehen, doch kommt sie wahrscheinlich auch in Sachalin und in Sibirien vor. Dieselbe ist eine nur zur palaearktischen Region gehörige Art.

12. **Chrysopa ogasawarensis** nov. spec.

Kopf gelb, rhombische Wulst auf dem Scheitel, Stirn und Clypeus orangegelb. Beide Taster hellorangegelb. Fühler etwas länger als der Vorderflügel, hellgelbbraun, nach der Spitze zu dunkler, das 1. und 2. grosse Basalglied orangegelb. Pronotum viel länger als breit, an den Seiten fast parallel, nach vorn zu gerundet, mit tiefer Querfurche fast in der Mitte der Länge; rauchgrün, mit einer gelben Mittellinie; fast unbehaart, doch an den Seiten fein behaart. Meso- und Metathorax ganz gelb. Abdomen gelbgrün, mit einer gelben Mittellinie. Beine hellgelbgrün, Schienen am äussersten Ende und die Tarsen hellgelbbraun.

Flügel am Ende zugespitzt, glashell, Pterostigma hellgelblich. Randmal, das gelbgrüne Basaldrittel ausgenommen, und die Adern ganz gelb. *Venulae gradatae* 5/8 bis 6/10 im Vorderflügel; 4/8 bis 5/8 im Hinterflügel. Kostalqueradern des Vorderflügels bis zum Pterostigma 21 bis 22; zwischen Radius und Radialsektor 12 bis 14 Queradern.

Körperlänge 12 mm.

Vorderflügelänge 16 bis 17 mm.

Hinterflügelänge 14 bis 15 mm.

Hab. Insel Bonin (Ogasawarajima), 2 Stücke am 20. August 1905 von Prof. S. Matsumura und zwei Stücke am 4. Juli 1912 von I. Kuwana gesammelt.

Diese Art steht *Ch. remota* Wk. nahe, ist jedoch durch die Thoraxfarbe deutlich zu unterscheiden.

13. **Chrysopa remota** Wk.

Walker, List Spec. Neurop. Ins. Coll. Brit. Mus., Pt. II, 1853, p. 238.

Hab.....Riukiu (Loo-Choo).

Sonstiger Fundort.Navigatorsinsel (nach Walker).

Leider habe ich diese Art noch nicht gesehen.

14. **Chrysopa formosana** Peters.

Petersen, Ent. Mitt., II, Nr. 9, 1913, p. 257.

Hab.....Formosa (Sokutso, Taihorin, Anping, Ako und Tappan).

In Formosa kommt diese Art sehr häufig vor, ausserhalb dieser Insel ist sie noch nie gefunden worden.

15. **Chrysopa ruficeps** Mac Lachlan.

Mac Lachlan, Tijdschr. vor Ent., 1875, p. 2, Pl. I, Fig. 1-4.

Hab. Formosa (Kankau, Giyochi).

Sonstige Fundorte. Celebes und Java.

Diese Art gehört zur orientalischen Region (Burma-China, Malayische und Celebische Subregion). In Formosa kommt sie verhältnismässig selten vor.

16. **Chrysopa basalis** Wk.

Walker, List Spec. Neurop. Ins. Coll. Brit. Mus., Pt. II, 1853, p. 239.

Hab.Riukiu (Loo-Choo Insel).

Ich kenne diese Art noch nicht.

17. **Chrysopa nipponensis** nov. spec.

Kopf dottergelb; Augen mit schmalem, blutfarbigem Saum, Wangen mit je einem schwarzen streifenartigen Fleck, hell blutfarbig, Clypeus an den Seiten mit je einem schwarzen Punkte, hell blutfarbig. Beide Taster gelb. Fühler kürzer als die Vorderflügel, dottergelb, an der Spitze etwas dunkler; das 1. dicke Basalglied an der Spitze hell blutfarbig

schmal gesäumt. Scheitel mit grosser, rhombischer, vorn zugespitzter, hinten abgerundeter Wulst. Pro-, Meso- und Metathorax oben mit einem dottergelben Mittelstreifen, hell blutfarbig, an den Seiten des Pronotums dunkler, *Scapulae anteriores* und Metanotum mit je einem dottergelben rundlichen Flecke; hinten gelb. Pronotum etwas kürzer als breit, nach vorn zu gerundet, mit tiefer Querfurche hinter der Mitte der Länge, fein behaart. Abdomen oben mit dickem, dottergelbem, an jeder Segmentspitze ziemlich breit ausgedehnt blutfarbig gesäumtem Mittelstreifen, dunkel-purpurn, hinten gelb; fein behaart. Beine gelb, nur Tarsen hellgelbbraun; gelb bis braun behaart.

Flügel mit etwas zugespitzter Spitze, glashell, Pterostigma hellgelbbraun. Randmal, Längs und Queradern gelb bis hellgelbbraun, Kostalqueradern in beiden Flügeln an ihrer Basis, Queradern zwischen Radius und Radialsektor in beiden Flügeln und zwischen Media und Cubitus im Vorderflügel an beiden Enden, Ramus des Radialsektors in beiden Flügeln und Ramus des Cubitus an der Basis und ein Paar der Basalqueradern des Vorderflügels, sowie auch *Venulae gradatae* schwarz. *Venulae gradatae* 6/7 bis 7/8 im Vorderflügel; 5/6 bis 6/6 im Hinterflügel. Kostalqueradern des Vorderflügels bis zum Pterostigma 22 bis 24; zwischen Radius und Radialsektor 11 bis 12 Queradern.

Körperlänge 8 bis 9 mm.

Vorderflügelänge 12 bis 14 mm.

Hinterflügelänge 11 bis 13 mm.

Hab. Hondo (2 Stücke in Kyoto, 2 in der Prov. Harima), Kiushiu
(2 Stücke in Kumamoto), Feb. bis Mai.

Gesammelt von W. Nakahara, M. Suzuki und Kawamura.

18. *Chrysopa bipunctata* Burm.

Burmeister, Handb. d. Entom., II, 1839, p. 982.

Hab. Hondo.

Diese Art wurde von Burmeister beschrieben. Seine Beschreibung

ist aber so einfach, dass ich nicht recht verstehen kann, auf welche Art sie sich bezieht. Deshalb habe ich sie nach der Beschreibung und Zeichnung von Schneider identifiziert. Ich habe nur ein Exemplar.

Dies ist eine sehr seltene Art.

19. **Chrysopa cognata** Mac Lachl.

Mac Lachlan, Linn. Soc., IX, 1867, p. 249.

Nothochrysa robusta Gerstaecker, Mitt. Ver. Neu-Vorpom. u. Rüg. Sg., 25, 1893, p. 73.

Chrysopa ricciana Navás, Revue Russe d'Entom., X, No. 3, 1910, p. 193.

Hab. Hokkaido (Sapporo, Jozankei), Hondo (Tokyo, Kofu, Provinzen Kii und Harima), Shikoku (Prov. Iyo), Kiushiu (Kumamoto, Nagasaki, Prov. Hiuga) und Formosa (Taihoku).

Sonstige Fundorte.....Sibirien (nach Navás); China (nach Mac Lachlan, Navás); Kambodscha (nach Mac Lachlan).

Diese Art ist sehr weit verbreitet und kommt häufig von Hokkaido bis Formosa vor. Ausser in Japan ist sie auch in Sibirien, China und Annam zu Hause, und zwar schwankt sie zwischen der palaearktischen und der orientalischen Region, was von grossem Interesse bezüglich der geographischen Verbreitung ist. Diese Species ist sehr ähnlich *Ch. septempunctata* (Wesm.) und auch *Ch. bipunctata* Burm. Unter den von Mac Lachlan zuerst untersuchten zahlreichen Arten gab es kein Exemplar, das einen schwarzen Punkt zwischen den Fühlern hat, wodurch sich diese Art hauptsächlich von *Ch. septempunctata* unterscheidet. Als er aber im Jahre 1875 die japanischen Chrysopiden-Arten bearbeitete, kam ihm ein Exemplar mit einem schwarzen Punkte zwischen den Fühlern zu Gesicht, worauf er diese Species als eine Lokalform der *Ch. septempunctata* bezeichnete, was ich aber nicht zugeben kann. Ich betrachte sie als eine selbständige Art durch den Kopffleck und die Farbe der Adern der Hinterflügel. Dieselbe unterscheidet sich auch sehr deutlich von *Ch. bipunctata* durch das Fehlen des sichelförmigen Fleckes vor den Fühlerinsetionen. Im Jahre 1893 hat

Gerstaecker zwei Chrysopiden-Arten beschrieben, von denen *Nothochrysa robusta* das Synonym von *Ch. cognata* ist. *Ch. ricciana* Navás (aus China) scheint mir mit dieser Art identisch zu sein.

20. *Chrysopa sauteri* Peters.

Petersen, Entom. Mitt., II, Nr. 9, 1913, p. 258.

Hab. Formosa (Tappan, Sokutsu, Kosempo), Hondo (Takasago).

Diese Art kommt in der orientalischen, sowie auch in der palaearktischen Region vor.

21. ***Chrysopa decorata*** Peters.

Petersen, Entom. Mitt., II, Nr. 9, 1913 p. 260.

Hab. Formosa (Kosempo), Hondo (Hiogo, Prov. Harima).

Petersen hat die Art von Formosa beschrieben. Es ist mir aber bekannt, dass sie auch im westlichen Teile von Hondo vorkommt. Dieselbe gehört zur orientalischen sowie auch zur palaearktischen Region, was für ihre geographische Verbreitung sehr interessant ist.

22. ***Chrysopa matsumurae*** nov. spec.

Grün bis grüngelb. Gesicht (vor der Fühlerinsertion) mit drei in einer Querreihe gelegenen schwarzen Punkten, von denen der mittlere fast rundlich ist, Stirn an den Vorderseiten mit je einem schwarzen streifenartigen Punkte. Clypeus mit zwei dunkelbraunen streifartigen Flecken. Oberlippe und Wangen schwarz gefleckt. Zwischen den Augen und Fühlerinsertionen ein kleiner schwarzer Punkt. Beide Taster hellgelbbraun. Fühler etwas länger als der Vorderflügel, hellgelbbraun, das 1. grosse Basalglied grün, aussen einen dunkelbraunen Streifen tragend. Pronotum viel breiter als lang, nach vorn sich etwas verjüngend. Mesonotum und *Scapulae anteriores* dunkelbraun. Beine hellgrüngelb bis blassgelb, Tarsen hellgelbbraun.

Flügel am Ende zugespitzt glashell, Pterostigma fast undeutlich.

Längs- und Queradern grün, die 1. Kostalquerader im Vorderflügel, die Kostalqueradern bis zum Pterostigma im Vorderflügel, an der Basalhälfte am oberen Ende, an der Apicalhälfte an beiden Enden, die Apicalhälfte des Hinterflügels, sowie auch *Marginales furcatae* in beiden Flügeln am Ende schwarz. In einigen Stücken sind die Queradern zwischen Radialsektor und Cubitus braun gesäumt. Vorderflügel mit zwei schwärzlichen Flecken, von denen ein grösserer sich am Basalviertel am Innenrande befindet, der andere an der Basis der 2. *Venula gradata*. *Venulae gradatae* 5/6 im Vorderflügel; 4/4 im Hinterflügel. Kostalqueradern des Vorderflügels bis zum Pterostigma 18 bis 19; zwischen Radius und Radialsektor 9 bis 10 Queradern. Adern struppig behaart.

Körperlnähe	6 mm.
Vorderflügelänge	9 — 10 mm.
Hinterflügelänge	8 — 9 mm.

Hab.....Hondo (Moji, Suma und Shizuoka), ziemlich häufig. Gesammelt von dem Prof. S. Matsumura, I. Kuwana und M. Shibakawa.

Gewidmet wurde diese Species dem ersten Sammler.

Diese Art steht der *Chrysopa decorata* Peters. sehr nahe.

23. *Chrysopa sachalinensis* Mats.

Matsumura, Jour. Coll. Agr. Tohoku Imp. Univ., Sapporo, Vol. IV, Pt. 1, 1911, p. 14.

Hab.Sachalin.

24. *Chrysopa nikkoënsis* nov. spec.

Grün. Wangen mit je einem schwarzen, nach innen gebogenen Streifen, zwischen den Fühlerinsertionen ein ziemlich grosser schwarzer Fleck. Beide Taster schwarz. Fühler so lang wie die Vorderflügelänge, die zwei grünen Basalglieder ausgenommen, hellgelbbraun. Prothorax breiter als lang, nach vorn sich verjüngend; oben mit sechs schwarzen Punkten,

von denen zwei in der Mitte sich befindliche fast rundlich sind, während vier an den Seiten streifartig erscheinen; schwarz, kurz behaart. Mesonotum mit je einem kleinen rundlichen, schwarzen Punkte. Beine grün, Tarsen hellgelbbraun; kurz, schwarz, dicht behaart.

Flügel am Ende etwas zugespitzt, glashell, Pterostigma ziemlich undurchsichtig und fast farblos. Randmal und Längsader grün, Kostalader an der Basis mit einem kleinen schwarzen Punkte, Kostalquerader bis zum Pterostigma in beiden Flügeln, Radialquerader in beiden Flügeln, Queradern zwischen Media und Cubitus in beiden Flügeln an beiden Enden, Cubitus- und Analqueradern in beiden Flügeln an beiden Enden, Ramus des Radialsektors an der Basis, und Verzweigungspunkt der *Marginales furcatae* des Vorderflügels, sowie auch *Venulae gradatae* schwarz gefleckt. *Venulae gradatae* 6/9 bis 7/9 im Vorderflügel; 5/8 im Hinterflügel. Kostalqueradern des Vorderflügels zum Pterostigma 25; zwischen Radius und Radialsektor 13 — 14 Queradern. Randmal und die sämtlichen Adern schwarz, kurz behaart.

Körperlänge 11 mm.

Vorderflügelänge 15 mm.

Hinterflügelänge 14 mm.

Hab.... Hondo (Ein Stück in Nikko, am 6. August 1913).

Gesammelt von Prof. S. Matsumura.

Diese Art steht *Ch. sachalinensis* Mats. und *Ch. cognata* M'L. nahe, ist jedoch durch den Fleck des Kopfes und Pronotums ganz verschieden.

25. **Chrysopa cognatella** nov. spec.

Diese Art steht *Ch. cognata* M'L. nahe, doch ist sie viel kleiner.

Grün, grüngelb oder blassgelb mit Ausnahme des grünlichgelben Thorax. Stirn an den Seiten mit je einem roten, kurzen und schmalen Streifen. Wangen mit je einem fast viereckigen schwarzen Flecke, ebenso Clypeus an den Seiten mit je einem kleinen schwarzen Flecke. Zwischen den Fühlerinsertionen ein schwarzer Punkt, der zuweilen fehlt. Beide

Taster schwärzlichbraun bis schwarz. Fühler etwas länger als der Vorderflügel, blassgelb, nach der Spitze zu dunkler; das 1. und 2. Basalglied hat aussen einen schwärzlichbraunen Streifen, der manchmal etwas undeutlich wird. Prothorax breiter als lang, nach vorn verjüngt und gerundet, hinter der Mitte mit tiefer Querfurche; oben mit zwei braunen bis rötlichbraunen Streifen. Beine blassgelb, Tarsen hellgelblichbraun, Klauen schwärzlichbraun.

Flügel am Ende etwas zugespitzt, glashell, Pterostigma ziemlich undurchsichtig. Randmal und Längsader grün bis blassgelb, Kostal- und Radialqueradern in beiden Flügeln, Ramus des Radialsektors in beiden Flügeln an der Basis, Queradern zwischen Media und Cubitus in beiden Flügeln (im Hinterflügel zuweilen an ihren Basen), und Verzweigungspunkt der *Marginales furcatae*, sowie auch *Venulae gradatae* schwärzlichbraun. *Venulae gradatae* 6/6 bis 7/7 im Vorderflügel; 6/5 bis 6/6 im Hinterflügel. Kostalqueradern des Vorderflügels bis zum Pterostigma 18 bis 21; zwischen Radius und Radialsektor 10 bis 11 Queradern.

Körperlänge 6 — 8 mm.

Vorderflügelänge 10.5 — 13.5 mm.

Hinterflügelänge 9.5 — 12.5 mm.

Hab..... Hokkaido (Sapporo, Jozankei, Ishiyama, und Iwamisawa), Hondo (Suma und Kyoto), Kiushiu (Kagoshima), im Mai bis September sehr häufig.

Gesammelt von Prof. S. Matsumura, W. Nakahara, M. Shibakawa und dem Verfasser.

26. **Chrysopa kurisakiana** nov. spec.

Diese Art steht *Ch. sauteri* Peters. nahe.

Grün bis grüngelb mit einer gelben Mittellinie. Wangen mit einem ziemlich grossen schwarzen Punkte. Clypeus an jeder Seite mit einem schwarzen Punkt. Beide Taster schwärzlichbraun bis schwarz. Fühler etwas kürzer als der Vorderflügel, hellgelbbraun, das 1. und 2. Basalglied etwas

heller. Prothorax länger als breit, nach vorn etwas verjüngt, hinter der Mitte mit tiefer Quersfurche, oben an den Seiten weit ausgedehnt dunkelbraun, und in der Mitte mit vier hellbraunen Punkten, die bei einigen Exemplaren manchmal undeutlich sind; dunkelbraun bis schwarz und lang (besonders an den Seiten) behaart. Beine hellgrüngelb, Schienen am äussersten Ende und die Tarsen hellgelbbraun, Klauen braun.

Flügel mit zugespitzter Spitze, glashell, Pterostigma hellgrün, etwas undurchsichtig. Randmal, Längsadern und Quersadern grün, *Venulae gradatae* schwarz, Kostalquersadern bis zum Pterostigma an ihrer Basis (im Vorderflügel) oder ganz (im Hinterflügel), Quersadern zwischen Radius und Radialsektor an der Basis (im Vorderflügel) oder an beiden Enden (im Hinterflügel) und einige Basalquersadern in beiden Flügeln, sowie auch bei einigen Stücken der Ramus des Radialsektors an der Basis dunkelbraun bis schwarz. *Venulae gradatae* 5/6 bis 6/6 im Vorderflügel; 5/7 bis 6/7 im Hinterflügel. Kostalquersadern des Vorderflügels bis zum Pterostigma 21 bis 22; zwischen Radius und Radialsektor 11 bis 12 Quersadern. Adern schwarz, lang und struppig behaart.

Körperlänge 8 mm.

Vorderflügelänge 13 mm.

Hinterflügelänge ca. 12 mm.

Hab..... Kiusiu (3 Exemplare in der Prov. Hiuga), Hondo (1 Stück in der Prov. Totomi, 2 Stücke in Tokyo, 1 Stück auf dem Berg Fuji, 1 Stück beim Towada-Sea bei Aomori).

Gesammelt von J. Kurisaki und Prof. S. Matsumura.

Gewidmet wurde diese Species dem ersten Sammler.



Geographische Uebersichtstabelle der japanischen Chrysopiden
 (Korea ausgenommen).

N a m e	Sachalin	Hokkaido	Hondo	Shikoku	Kiushiu	Riukiu- Insel	Formosa	Sonstige Verbreitung
Nacaura matsumurae					+			
Nothochrysa japonica			+	+	+		+	
N. olivacea			+					
Ancylopteryx octopunctata							+	Cochinchina, Indien, Java, Ceylon, u. Philippinen.
Pseudochrysa formosana							+	
Chrysopa perla			+					Europa, Sibirien
C. nigriceps			+					
C. intima	+	+	+					Sibirien
C. lezeyi		+	+					
C. sapporensis		+	+					
C. furcifera			+		+	+	+	
C. anpingensis							+	
C. boninensis			Insel Bonin*				+	
C. vulgaris var. microcephala			+					Europa
C. inornata	+		+					
C. vittata		+						Europa
C. ogasawarensis			Insel * Bonin					
C. remota						+		Navigatorsinscl
C. formosana							+	
C. ruficeps							+	Celebes, Java
C. basalis						+		
C. nipponensis			+		+			
C. bipunctata			+					
C. cognata		+	+	+	+		+	Sibirien, China, Cambodja
C. sauteri			+				+	
C. decorata			+				+	
C. matsumurae			+					
C. sachalinensis	+							
C. nikkoënsis			+					
C. cognatella		+	+		+			
C. kurisakiana			+		+			
T o t a l	3	6	21	2	7	3	11	

* Die Insel Bonin gehört eigentlich zur subtropischen Region, aber sie ist wegen ihrer Lage bequemlichkeitshalber in Hondo eingeschlossen.

P. S. Nach der Abhandlung „Les Chrysopides du Musée de Londres“ (Annales de la Société Scientifique de Bruxelles, XXXVIII, 1913—'14, Premier fasc.) welche ich während des Drucks dieser Arbeit bekam, findet sich *Chrysopa formosa* Brauer auch in Japan, worauf ich hingewiesen haben will, obwohl ich dieselbe noch nicht gesehen habe.

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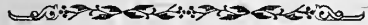
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OBSERVATIONS ON HOTARU-IKA WATASENIA SCINTILLANS.

By

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Tohoku Imperial University, Sapporo, Japan.

(With 3 plates and 1 text figure.)

PREFACE.

I had an opportunity to observe the living state of *Hotaru-ika* **Watasenia scintillans** (*Berry*) and the actual scene of fishing for them at Namerikawa, Toyama Prefecture in 1913. I was there twice in that year, first in spring from April 17 to May 10, and again in summer, from July 21 to August 23.

The present investigation embodies the studies concerning mainly the habits of the *Hotaru-ika*. The observations on their living state as well as the references to the fishery were made in the visits mentioned above¹⁾ and the materials used in this study consist of the specimens obtained there as well as of those from other localities.

The fishing season of *Hotaru-ika* in Toyama Bay and its coasts is commonly late April and the whole of May. The coast where this fishing is carried on, extends from Ôta, Himi-gun, Etchu Province to Itoigawa, Echigo Province, which forms the deepest inlet of the Japan Sea in Honshu.

1) I express my warmest thanks to Mr. K. Koishi, Principal of the local Fishery Institute, Namerikawa, and his staff, especially Mr. S. Matsuno, for their efforts in helping me in every way in my investigation.

The total amount of the annual catch reaches generally about 1000 tons, though it shows some fluctuation, and in 1913 it reached a little above 900 tons, the statistics here being taken directly from the notes of fishermen engaged in that fishery. *Hotaru-ika* caught there are mostly mature females; young ones have not been found.

For facilities and useful advice given me in the present work, it is my pleasant duty herewith to return my cordial thanks to Prof. Dr. T. Fujita.

I. HABITS OBSERVED FROM THE ZOOLOGICAL POINT OF VIEW.

1. Sexual Dimorphisms.

The characteristics of the female *Hotaru-ika* have been already made public by S. S. Berry (Proc. Acad. Nat. Sci. Philadelphia, 1912, p. 425), and the specimens examined by me practically agree with his description except in details, which do not seem sufficiently important to be stated here.

The principal sexual differences based on the study of 20 formalin specimens of each sex obtained in the period from April 25 to May 7 are as follows:

Female.—The mantle-length is 54–67 mm. This is about the minimum and maximum limits of the mantle-length of the fully mature female, for I have never met with any beyond these extremities.

Two-sevenths of the mantle at the anterior is cylindrical. From thence it tapers gradually and is pointed at the posterior end. The fin is large and very broad, the breadth being distinctly greater than the length, which is slightly less than $\frac{2}{3}$ of the mantle-length (Pl. I, fig. 1).

The pen is of the *Loligo*-type and wide, the breadth being a little less than $\frac{1}{5}$ of the length (Pl. II, fig. 1).

Male.—The body is a little smaller than that of the female, the mantle-

length being 44-54 mm.; this may also be taken as the length of the fully mature male for the same reason as stated above. The mantle is slenderer than that of the female and rather conical in shape, the broadest part being in the anterior. The fin is of equal breadth and length, and is proportionately smaller than that of the female, the length being only a little longer than half the mantle-length (Pl. II, fig. 4).

The hectocotylus is the right ventral arm, and provided with 2 small semilunar membranes in the terminal portion, but beyond that it shows no special modification (Pl. II, fig. 3).

The pen is distinctly narrower than that of the female, the breadth being about $1/7$ of the length (Pl. II, fig. 2).

2. Luminous Organs and their Phosphorescence (Pl. II, figs. 2-5).

Prof. Dr. S. Watase was the first to make zoological observations on the phosphorescence of the luminous organs of *Hotaru-ika* at Namerikawa, and he described it in the *Dôbutsugaku Zasshi*, Tokyo (1905, p. 119), and also announced it at the Seventh International Zoological Congress at Boston, U. S. A., 1907. The luminous organs are classified here also into 3 kinds according to Prof. Watase, i. e. (a) brachial organs, (b) minute organs scattered on the ventral surface of the whole body, (c) ocular organs.

Brachial Organ. This is the largest organ, and when I made observations in the fishing season, it was much more active in phosphorescence than other organs. It is situated at the end of each ventral arm, composed of 3 globules arranged in a series. The globules are ovoid in shape and nearly squal in size, but the middle one in the series is generally a little larger than the others, the dimensions being 1.4 mm long and about 1 mm broad. In fresh specimens they show a greenish cobalt colour, and there are 2 or 3 layers of large brownish chromatophores covering a part of the preceding substance (Pl. I, fig. 4). These chromatophores are constantly contracting and expanding. When they were observed at night on the living animals, they were seen to discharge light in all directions much brighter than any of Japanese fireflies. The colour of the light is

Prussian-blue or tinged a little with purple, and the luminosity is strong enough to absorb the light of other luminous organs. When the living animal was placed on a glass plate, which was put directly on the case of the dry plate of the photographic camera, and then exposed for 4 seconds with the Lion's dry plate of the special rapid no. 230, the light of this brachial organ was distinctly taken on the dry plate, although those of other organs made no impression.

Minute Organs Scattered on the Ventral Surface of the Whole Body. There are numerous minute organs distributed on the ventral surface of the mantle, head and siphon, and they are also on the third and fourth arms. The external appearance as well as the histological character is the same but with some diversity in size.

The organs on the mantle differ in number between male and female, i. e. 566-687 in the female and 450-543 in the male as counted in 10 specimens of each sex. They are entirely found on the ventral half of the mantle, and the arrangement is not quite regular, but more or less in series showing slight bilateral symmetry. The distances between the organs are nearly equal but with slight gradual increase towards the lateral sides as well as towards the posterior end of the mantle.

The organs on the siphon are about 47 in number, alike in male and female, and the arrangement resembles those of the mantle.

The organs on the head are about 180, alike in male and female, arranged somewhat in several series with the anterior organs of these series connecting with the series of the same organs on the arms. Besides, along the margin of each eye-lid there is another series, which consists of larger and smaller organs arranged alternately.

The organs on the fourth arms are arranged in 3 series, the middle one of which is the longest, consisting of about 28 organs, and connecting distally with the brachial organs, beyond which yet 3 other small organs are found. The brachial organ from the point of view of situation appears to have evolved from the minute organ but of course with great

histological differences. The third arms have only about 7 organs arranged in a series.

Each organ in the fresh specimen has a substance of purplish hue in the centre; this substance seems to be that discharging light when the animal is living. When the organ is exposed in the air, the purplish hue of the substance changes to greenish blue after a while, and finally resolves into a true green (Pl. I, fig. 3). The substance is covered by a pigment layer of darkish brown or deep purple which has a hole resembling the pupil of an eye, through which the substance can easily be seen. The light of the substance at night is whiter and less luminous than that of the brachial organ.

Ocular Organ. When the eyelid of the fresh specimen is removed and the eyeball exposed, there are seen 5 luminous organs arranged in a series along the ventral circumference of the eyeball, the organ on either end of the series being a little larger than the remaining 3. The colour of all these organs is pearly white (Pl. I, fig. 5). When the organ is seen at night in the living animal, the phosphorescence is not distinguishable from that of the minute organ on the body.

Difference of Phosphorescence in the Sexes. On examining the preserved specimens to discover the difference of the external forms as well as the histological structures of their luminous organs as occurring in the male and female, none could be discerned. But in the female specimens there are one hundred or so more of the minute organs of the mantle than in the male. Whether there is any meaning as to sexual selection, it is difficult to say, the data concerned being insufficient at present to announce any opinion.

Next, as to the difference of phosphorescence between the sexes in their living state, the means of investigation proved to be very difficult. At first I repeatedly undertook to keep the animal in an aquarium, but no success was attained. The reason for the failure is that first of all, the animals are very delicate, and next the aquarium was defective. The

animals are so weak that in carrying them from the sea to the aquarium they wasted and died. As they wasted, the luminosity in question became very feeble, and naturally with their expiration, the light of the luminous organs gradually vanished altogether. This being so, I then tried to observe the animals directly while they were swimming in the net. But no good means were found easily to distinguish the sexes on such dark nights, even with the feeble light of the moon or of a lantern.

However in my examinations at night, no special variety of the light could be found, the colour of the light being always the same. And in one case, putting in a vessel and observing about 30 specimens in a fishing boat while they were yet actively on motion, I verified the fact that their luminosity is uniform. In the morning, to my surprise, a male was found dead among those 30 specimens; this proves that it had the same colour of light with the female on that night. The above data seem to prove the fact that the colour of the light of the luminous organs is the same in both sexes.

Again, in late July of the same year, I made another observation on the phosphorescence under consideration and then it was quite evident to me that the luminosity of the brachial organ was at this season noticeably feebler than in the spring.

The phosphorescence of the immature animal can never be studied in Namerikawa, young ones thus far not being found there.

3. Food.

The contents of the stomachs were examined 4 times on the preserved specimens and they were as follows:

- i) Among 20 females caught with the *Fukube-ami* (see p. 91) on April 25, were 4 specimens with some amount of silvery blackish substance like the iridocytes of fishes and some pieces of shells of small crustaceans, but the remaining 16 had nothing in the stomach.
- ii) In 20 females obtained with the same kind of net on April 27, the stomach was found to be quite empty.

iii) In 6 specimens out of 50 females caught with the same net, April 29, the stomachs contained rich and rather fresh contents, consisting mainly of small fishes, *Mysis* sp. and pelagic Copepods¹⁾ (*Corycaeus* sp. & *Oncaea* sp. etc.).

iv) 3 specimens out of 10 females obtained, July 26, had some iridocyte-like substance, some pieces of the back bone of a fish and 2 pieces of its skull.

Besides, there were also found in the stomach some plankton-animals which were indistinguishable on account of their maceration.

Judging by the facts stated above, it seems that the animals having food in the stomach are few in percentage, and we may say that the *Hotaru-ika* caught during the night at Namerikawa and vicinity are not of those kinds which visit the sea coast for the purpose of getting food, i. e. feeding migration.

4. Seasonal Changes of Liver Content.

The fresh female *Hotaru-ika* in the fishing season are at a glance distinguishable from other kinds of cuttle-fish, being reddish in colour. This colour is caused by the liver, which is seen externally through the transparent mantle. If the liver be dissected in water, many reddish oily globules (reserved nutriment?) come up to the surface of the water. And if the liver be split into small pieces with a dissecting needle and looked at under the microscope, it is easily seen that the same reddish substance is also contained in the tissue. But in the males which were taken at the same time with the females, the liver shows no such reddish hue, but is gray and provided with only a small amount of such reddish oily substance in the tissue. This difference in colour between the sexes is so easily distinguishable in the spring time that above 10,500 specimens were assorted into male and female by 5 or 6 students of the Fishery Institute in about one hour and a half.

But, on my second visit, which lasted from the middle of August till late September, it was soon observed that the reddish hue of the liver of the female was changed into a gray hue resembling that of the male in spring. The

1) I owe the identification of the Copepoda to Mr. S. Kokubo, to whom I express my thanks for it.

consistency of the liver at this season is soft, and the oily content is poor as compared with that of the reddish-hued liver in the spring, which is thick and rich.

Besides the diminution of the oily substance in the liver, all the organs generally seem to undergo degeneration, and the vitality of the animals is considerably lessened. They expired soon even with gentle handling, and though they were caught by hand they were very quiet, while in the spring time they attacked us violently, biting our hands with their jaws. From the middle of July, the catch of *Hotaru-ika* became gradually less and less, until in August and September, a net which had in the spring fishing season caught about 500 pounds at a time caught only a very small number. Later it became yet smaller, and very feeble animals only were occasionally trawled from the depth of 100 or more fathoms.

By the above facts, we are justified in saying that they probably die with the passing of summer.

5. Female Genital Organ.

When the mantle of the female caught with the *Fukube-ami* was dissected along the ventral median line, it was seen that its posterior half is filled up with many transparent small eggs of fairly greenish hue.

Most of the eggs are mature and are freely contained in the ovarian sac, which is divided into 2 parts by a median septum. And a comparatively small number of immature eggs are imbedded in the ovarian tissue. From the central portion of each half of the ovarian sac there arises an oviduct with a thin and transparent wall. This oviduct runs backward along its surface till near the posterior end of the sac. Then it turns forward, and opens to the nidamental gland attached at the anterior end of the sac. In this season the oviduct is found to contain a quantity of the fully mature eggs (Pl. II, fig. 10).

The nidamental gland is a slightly pinkish body of inverted V-shape situated behind the siphon; each ramus of the gland is split dorso-ventrally into 2 lobes with the exception of its posterior $1/6$ part. The oviduct arrives

at the posterior end of this ramus, and piercing through the matrix of the gland it finally opens to the posterior end of the inner face of the lobe. The inner face of the lobes are divided into numerous fine lamellae and secrete some albuminous substance (Pl. II, fig. 11).

Judging from the above facts, it seems that the eggs when mature, are finally discharged between the lobes of the nidamental gland, whence they are covered with albuminous substance secreted from the lamellae thereon. Then they are dispelled through the siphon.

6. Number of Ovarian Eggs of the Female.

The number of the mature eggs was reckoned from the females which were directly obtained from the net, on the ground that otherwise the specimens might have sustained some harsh treatment and so have lost some of their eggs. By this count, it was found that the number of mature eggs varies in the different individuals, and the largest number is about 1200. The actual numbers of the mature eggs of 25 females caught at the same time, together with the numbers of spermatophores fixed by the male on both sides of the nape of these females, are shown in the following table :

Characters Specimens	Mantle-length	Approximate number of eggs	Number of spermatophores	
			left	right
	m m.			
1	68.4	730	7	6
2	67.6	960	7	7
3	67.0	950	7	7
4	67.0	950	5	4
5	65.5	1,200	5	7
6	65.5	850	5	6
7	65.0	1,080	6	7
8	64.0	1,050	8	8
9	63.0	1,100	1	6
10	63.0	850	6	8
11	63.0	600	7	3

Characters Specimens	Mantle-length	Approximate number of eggs	Number of spermatophores	
			left	right
	mm.			
12	62.5	720	4	12
13	62.5	980	7	5
14	62.0	960	8	7
15	60.8	1,200	3	3
16	60.5	950	6	5
17	60.0	480	6	6
18	58.5	470	7	6
19	56.8	1,070	8	7
20	55.6	730	9	5
21	55.0	850	5	1
22	53.0	860	5	3
23	53.0	720	8	9
24	52.0	950	6	8
25	46.0	730	8	8

Such a diversity in the number of the mature eggs according to the individuals is attributable either to their premature condition or to their discharged effect, but here the latter cause is more reasonable to explain the fact. That is to say, the diversity is due to the fact that they are in the midst of the spawning season.

7. Male Genital Organ.

The anatomical relation of the male genital organ does not differ on the whole from that occurring generally in the Enoploteuthidae. The number of the spermatophores in Needham's sac differs according to the individuals, and there are 73 in the fully extended sac of a male.

The spermatophore (Pl. II, fig. 7) examined under the microscope shows a delicate organ covered with an outer sheath. This organ consists of 3 parts: a packet of spermatozoa which fills nearly 1/10 of the outer sheath of the spermatophore (Pl. II, fig. 7, A); a long elastic spiral spring (ibid. fig. 7, B); and a tubular sheath connecting the preceding 2 parts (ibid. fig. 7, C). And

as soon as the spermatophore is stimulated with the point of the dissecting needle, the spring and connecting sheath coil together forming a ball-shape. By this action the packet of the spermatozoa moves toward the other extremity, and at the same time, the outer sheath of the spermatophore breaks up. Now the packet comes out, and by its rupture the spermatozoa are discharged therefrom (Pl. II, figs. 8, 9).

8. Spermatophores Fixed in the Female.

All the females which were caught from the latter part of April till the middle of May had without exception 2 bundles of spermatophores, so far as I am aware, which were tightly fixed on the nape under the velum of the siphon. Here it has a special adjustment in a form of a pit so as to receive the bundles of spermatophores (Pl. II, figs. 5,6). The number of spermatophores in a bundle differs with the individuals as well as with the bundles of each individual. This number varies from 1 to 13 but as a rule it is about 7. This was ascertained from 50 specimens caught at the same time.

The spermatophores show no greater variation in their number than that just described, even in the specimens of the later seasons. Here it must be emphasized that the female individuals with spermatophores fixed have no new addition by other males. The spermatophores are seen always to fuse together in a bundle at the basal extremity, thus proving that they were fixed at one time. Each spermatophore of an attached bundle shows the same configuration as those treated artificially as mentioned before.

9. Facts Concerning Fate of the Male.

Discovering, from the fresh specimens which I obtained at Namerikawa, that every female has a more vividly coloured liver than the opposite sex and in this season always carries 2 bundles of spermatophores at its nape, I showed these differences to some students of the Fishery Institute, who henceforth were able easily to select 132 male specimens from about 10,500 specimens of the total catch on April 25, 1913. In like manner, many males were obtained as given in the following tables, which show at the same time the ratio of the male to the female.

Table I : *Hotaru-ika* Caught with *Fukube-ami* at Night.

	Total number of specimens	Number of males	Ratio of males to females	Locality	Date
I	cirea 10,500 (calculated from the weight 282 lb, reckoning one piece as 0.43 oz)	132	1 : 78.8	Namerikawa	April 25
II	cirea 5,333	26	1 : 204.1	do.	April 30
III	282	1	1 : 281	do.	May 4
IV ¹⁾	cirea 12,326 (calculated from the weight.)	10	1 : 1,231.6	do.	May 28
V ²⁾	cirea 15,550 (calculated from the weight.)	19	1 : 817.4	do.	May 29

The above table shows clearly that the male becomes rarer as the season advances.

Table II : *Hotaru-ika* Gathered with a Drag-net³⁾ during the Daytime at a Depth of 100 or a few more Fathoms.

	Total number of specimens	Number of males	Ratio of males to females	Date
I	3	1	1 : 2	April 24
II	653	3	1 : 216.7	May 2
III	85	2	1 : 42.5	May 2
IV	52	1	1 : 51	May 3

Table III : *Hotaru-ika* Obtained from the Stomachs of *Theragra chalcogramma* (Pallas).⁴⁾

	Number of fish	Total number of <i>Hotaru-ika</i>	Number of males	Ratio of males to females	Locality	Date
I	10	5	0	0 : 1	Namerikawa market	April 19
II	30	6	0	0 : 6	Off Ikuji	April 23
III	30	7	1	1 : 6	Namerikawa market	April 30

1); 2) found out by Mr. S. Matsuno.

3) This net is used for fishing for *Nigisu* (*Argentina semifasciata* Kishinoue).

4) *Theragra chalcogramma* is caught with the line from the depth of 100 or a few more fathoms and in the sea 3 or more miles off the coast.

The preceding tables clearly show that the male is less in number than the female whether it was caught with *Fukube-ami* at night, or with the dragnet during the day, or obtained from the stomach of fish.

From the preceding pages we get the following data: 1) the number of spermatophores comprised in a fully extended Needham's sac is about 73 (see p. 84); this number may be reckoned as the total sum of the spermatophores produced by a male in a year for such an animal finishing its mating in a short period, 2) the number of the spermatophores fixed on the nape of the female is 14 on an average, for each female has 2 bundles of spermatophores, each comprising 7 ones on an average (see pp. 83, 85). If we assume that all the spermatophores of the male are entirely fixed into the females, then it is necessary at least that there should be one male to about 5 females, considering from the above two facts. But Table I on page 86 shows that the ratio of the males is less than one against 79 of the females so that there are in the animals caught by the *Fukube-ami* at night, only $1/5$ enough males to suffice for mating. So too in the specimens obtained either from the deep of the sea or from the stomachs of the *Theragra*, we get the same tendency in the numerical relation of the two sexes.

Further, there may be some mistake as to the fixation of the spermatophores by the male. Consequently the enumeration of males by the number of spermatophores becomes uncertain and many more of them should naturally be expected.

10. Eggs Floating on the Sea.

The egg is ovoid in shape, being 1.5 mm. long and 1.2 mm. broad. Those obtained between April 19 and May 7, 1913, showed either the segmented condition or a further developing stage. The sea condition at the time of the surface collection of the egg is as follows:

Date	Beginning of collection	Duration of collection	Condition of sea	Specific gravity of sea water	Number of eggs
1913 April 19	X a. m.	50 minutes	calm	1.0152	2
April 20	VI 1/2 a. m.	1 hour	calm	1.0165	11
April 22	VI a. m.	40 minutes	calm	1.0150	3
April 29	I a. m.	1 hour	calm	1.00695	0
May 2	X a. m.	1 hour	calm	1.0111	0
May 6	VI a. m.	1 hour	calm	1.0144	21
May 7	IX a. m.	1 hour	calm	1.0154	12

The repeated investigations made by Mr. S. Matsuno had verified the fact that the eggs float in greatest numbers on the sea surface a little more than half a mile off the coast. And the season in which the eggs are collected most thickly on the surface agrees with the fishing season along the coast of Toyama Bay. The eggs obtained early in the fishing season show an earlier developmental stage, thus confirming the opinion that the littoral migration of this animal is simply for the purpose of spawning.

11. Conclusion.

Hotaru-ika caught in enormous quantities in Toyama Bay have the following habitual characters:

- a. A large percentage of them belong to the female sex; these are sufficiently matured, carrying spermatophores fixed by the male.
- b. There should be many more males than are caught in spring.
- c. The littoral migration is for spawning.

After the preceding investigation, I examined many specimens of *Hotaru-ika* from Sagami Bay preserved at the Zoological Institute, Science College, Imperial University, Tokyo, and found from them many interesting facts which seem to verify my preceding opinions. These specimens in the order of seasons caught are arranged as follows:

- i) 81 specimens,—all matured males, captured at Shirahama, Awa Province, Feb. 9, 1928. The mantle-length being measured from 12 out of the whole number varies from 36–45 mm.

ii) 4 specimens,—all mature males, donated by Prof. Dr. A. Oka who obtained them at Hazama-mura, Awa Prov., in the middle of Feb., 1907. The mantle-length, 36–42 mm.

iii) 16 specimens,—all mature males, obtained at Nishimisaki, Awa Province, Feb., 16, 1891. The mantle-length, 36–44 mm.

iv) 104 specimens,—♂ 37 + ♀ 17, Odawara, Sagami Province, March 9, 1907. The males are here also all matured, and the number of spermatophores carried by the females is as follows :

Female specimens Characters																	
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
Mantle-length in mm.	37	39	39.5	40	40	40	41	41	41	41.5	42	42	42	42.5	44	44	45
spermatophores in nape	right	0	0	7	0	0	5	7	8	0	7	7	4	6	0	7	8
	left	0	0	0	0	0	5	7	5	0	4	7	5	8	0	4	8

v) 33 specimens,—♂ 10 + ♀ 23, Odawara, Sagami Province in the middle March, 1900. The males are matured, and the females, except only one, carry numerous spermatophores.

vi) 4 specimens,—all mated females, caught by Mr. H. Ôshima, at Manazuru, Sagami Prov., March 18, 1909. The mantle-length, 45–49 mm.

The above specimens convince us that on the coast there exist numerous males especially in the early fishing season, that diminish in inverse proportion with the female as the season advances.

The first females caught do not carry any spermatophores on the nape, i. e. are unmated. Such specimens constitute about 35 % of all the females in the fourth bottle, and about 3.4 % in the fifth bottle, thus the unmated females diminish gradually in that ratio; this seeming to show that all the females that come to spawn are unmated at first. So in Suruga Bay the mating of this animal probably takes place in early March and the spawning probably in April.

By these differences in the mating and spawning habits of the same animal

but living on the opposite sides of the Empire I am convinced that these different modes of reproduction are in reality a single and continuous process; this means that passing through such a habitual progress in Sagami Bay, the animal arrives at the condition of its life at Namerikawa and vicinity.

The fact that only unmated females come to spawn naturally means that those once mated never come. This seems to confirm my opinion that they probably die as the summer passes (see p. 82). The male probably dies after the fixing of spermatophores in the nape of the female, and the few lately matured ones are caught in company with the female in *Fukube-ami* in April and May.

II. HABITS OBSERVED FROM THE FISHERY POINT OF VIEW.

Methods of Investigation.— The statistics of the annual catch were derived from the daily notes of fishing of the persons who are carrying on the *Hotaru-ika* fishery, also from information given by the fishermen who were directly doing the fishing. The maps of the fishing places with their depth were made according to the following method:

- i) Maps published at the Hydrographic Office, Naval Bureau, as follows:
 - a) Main part of Japan and Korea.
 - b) Tsuruga Wan to Niigata Kô.
 - c) Noto Peninsula.
 - d) Fushiki and Yuwase Anchorage.

The isobathymic lines and coast-lines in this paper are mainly dependent upon the preceding maps, their details being referred to the maps and to notes of the fishing places, according to which fishermen fix the nets, as well as upon the soundings taken by myself.

- ii) Maps of the fishing places used for Fishery Legislation in the Toyama Prefectural Office.

The situations of *Fukube-ami* in the maps contained in this paper are drawn from these maps, some corrections being made through the kindness

of the influential persons carrying on the *Hotaru-ika* fishery as to the situations of the nets in 1913.

A. FISHING CONDITION OF THE WHOLE COAST.

The coast-line of Toyama Prefecture is curved in and embraces Toyama Bay. It is very deep along the whole coast, the 100-fathoms line being nearer the shore than anywhere along the coast of the Japan Sea. And the middle portion of Toyama Bay has a depth of above 600 fathoms.

The sea bottom inclines generally very irregularly at the coast and there are many valley-like places near the sea-shore, this being especially the case at Namerikawa, while the bottom consists generally of darkish gray mud. The tidal current on the coast is a tributary of the Tsushima Current and comes from the Noto Peninsula, running toward the Province of Echigo. The surface temperature of the sea water in the *Hotaru-ika* fishing season is about 10°C at the beginning, increasing to about 14°C at the most prosperous season of the fishing, and reaches 18°C towards the latter end.

Hotaru-ika are obtained very frequently in spring from the depth of 100 or more fathoms in the sea, about 2-7 miles off the coast, proving that at least some part of them are living in that region during the fishing season.

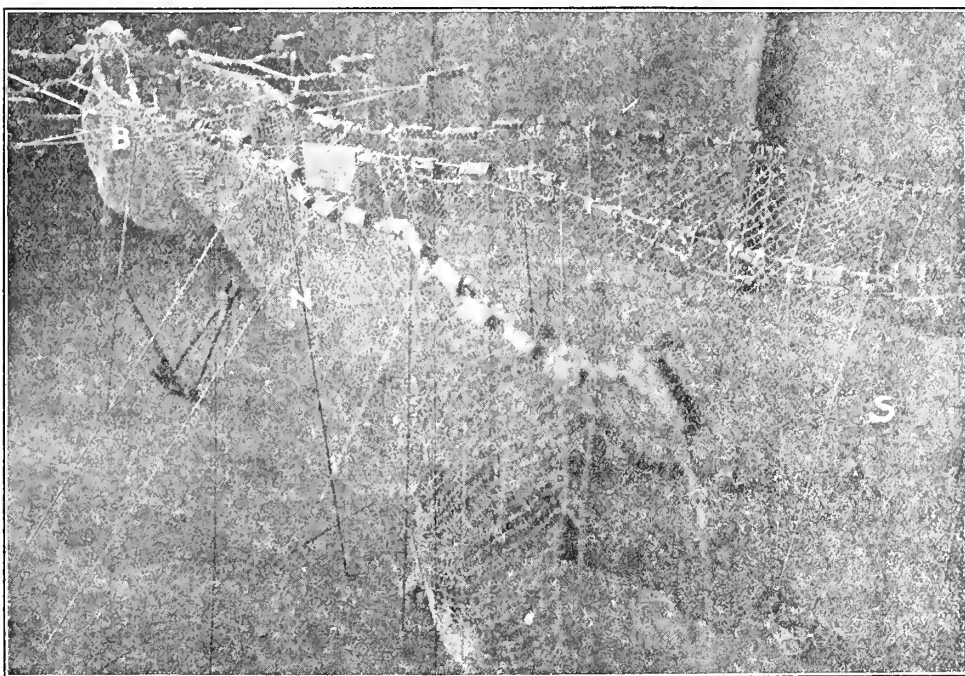
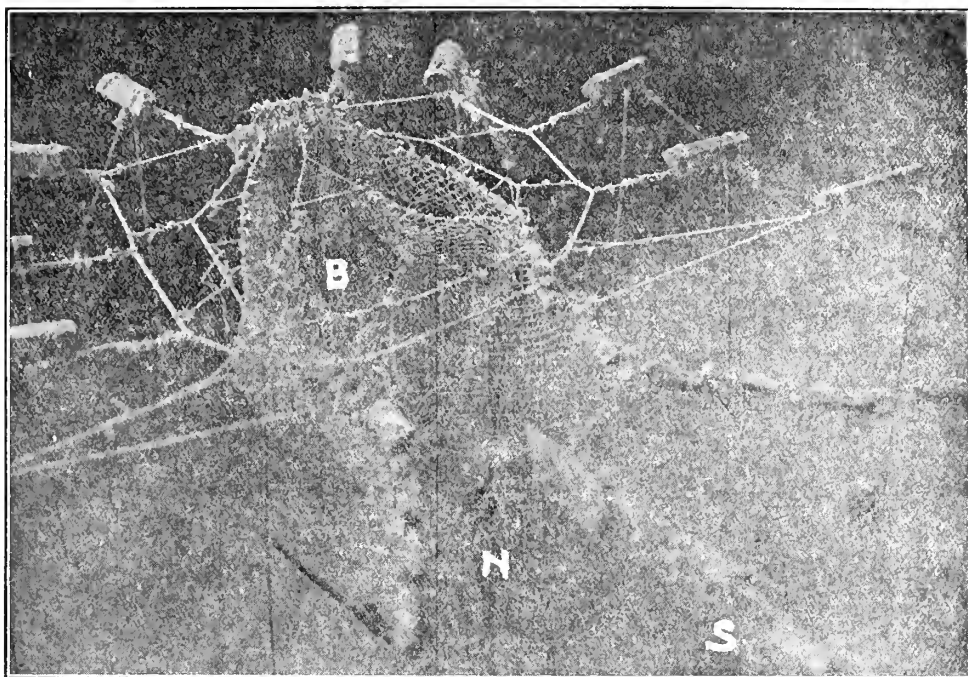
The most useful fishing implement used by the fisherman for the *Hotaru-ika* fishery in the Toyama Bay is the "*Fukube-ami*". On the coast of Toyama Prefecture, the *Fukube-ami* is generally fixed sometimes in ranges on the steep slopes of the valley-like depressions or on the submarine banks; these stand at right angles to the shore-line.

The *Fukube-ami* is made of 2 principal parts, i. e. a bag-net and a screen-net. The screen-net is about 20 meters in extent and is fixed in the same direction as that of the submarine valleys or banks to cut off the way of the fish driving them into the bag-net toward the open sea, its under edge being generally fixed into the sea bottom. The bag-net is fixed at the extremity of the screen-net, and has a funnel-like mouth opening toward the shore, the

bottom of the funnel having an inclination which becomes lower toward the shore, fixed with its edge into the sea bottom as in the case of the screen-net. Therefore the fish, even those moving near the sea bottom, being driven in this direction, run on the funnel and are entrapped in the bag-net. The *Fukubi-ami* has many forms, but it is only necessary to mention two in this paper. The first is made with a double opening, the bag-net has a mouth on both sides of the screen-net so that it entraps the school equally from both sides. This kind is used for fixing on the back of narrow banks with the deep sea at both sides. The second is made with only one opening,—the bag-net has a mouth on only one side of the screen-net, so that the school comes into it only from the mouth-bearing side. This is used for fixing on the slopes of these submarine valleys, which have a depth only at one side and a shallow vast floor on the other.

The schools of *Hotaru-ika* come into the net in the fishing season after sunset and never during the day, and the net drawn up at 9 or 10 p.m. is better filled than that drawn up at 3 or 4 a.m. This fact seems to suggest that the schools come near the coast from the deep in the offing when sunset approaches, and as soon as they have laid the eggs towards evening go back in to the deep sea, and are entrapped in the net on their way back. They come very rarely to the surface of the sea even at night.

There are 2 or 3 special submarine valleys of 100 or more fathoms, where various deep sea animals are caught, for example, *Nigisu-sebikiba* at Uozu (Pl. III, fig. 5; p. 86, footnote) and *Hirataebi-teguri-amiba* at Shimminato (ibid. fig. 2; p. 93). In those places, even in the daytime in the fishing season, *Hotaru-ika* are always gathered by a drag-net, though in other shallow parts, they are never caught. So it suggests that in such a depth even near the sea coast, they live also in daytime. And there are also several places where in the fishing season *Hotaru-ika* are gathered by a hauling seine from land, after sunset, for example, Takatsuki at Namerikawa (ibid. fig. 4), *Tai-jibiki-ba* at Uozu (ibid. fig. 5) and *Baichi* at Kyoden (ibid. fig. 6), where there seem to be some suitable slopes for *Hotaru-ika* to approach to the coast, having a vast



Two views of distal part of *Fuku-be-ami* (model). B: Bag-net, N: funnel like part of bag net, S: screen-net.

valley very near.

Those facts show that *Hotaru-ika* is a deep-sea animal, living during the day in a depth of 100 or more fathoms, and when the night is at hand, they approach to the coast, and after sunset they lay the eggs, and as soon as they finish their spawning, go back to the deep sea.

B. FISHING CONDITION IN LOCAL PARTS.

The whole coast of Toyama Prefecture is divided for convenience's sake, into 3 regions,— i. e. Shimminato region, Namerikawa region and Ikuji region.

1. Shimminato Region.

This consists of the western part extending from Yokata along the whole coast of Toyama Prefecture. The 100-F-line is the farthest off the coast in all that region, especially in Himi-gun, where the distance from the shore is about 3 or more miles generally, and the bottom of this coast shows a more gradual slope than that of others.

This part has two principal submarine valleys coming near the shore, the first being that off Shimminato and Fushiki, and the second that off Yokata. And the greatest number of *Fukube-ami* are settled at the boundary walls of those two valleys and the nets which catch *Hotaru-ika* in great quantities also belong to those *Fukube-ami*. This region is again divided into 3 coasts.

a) *Shimminato coast* (Pl. III, fig. 2). This coast is the central part of the whole and has 3 rivers, i. e. Shô river, Shinshô river and the river flowing from Hôjôzu Lake, and there is, in front, the valley off Fushiki and Shimminato, the central part of which showed about 170 fathoms by my sounding. In the spring, *Hotaru-ika* are gathered during the day with a drag-net which is used for catching the *Hirata-ebi*, a deep-sea shrimp, from a depth of 100 or more fathoms.

The following table shows the amount of *Hotaru-ika* caught with *Fukube-ami* along this coast in 1913.

Situation of net	Number showing net in map	Name of net	Direction of net to tidal current	Catch
				Kan (= 8.26 lb +)
Off Ebie	—	Senaka-niban	Following	800
do.	—	Naka-hōji	Against	640
do.	—	Nawatake	Against	800
Off Horioka	1	Ikirei	Against	748
do.	2	Dairaiboku	Against	368
do.	3	Kiawase-segata-samban	Against	520
do.	4	Kiawase-segata-niban	Against	464
do.	6	Higashi-samban	Following	300
do.	5	Kaguradashi	Following	400(1912)
Off Shimminato	8	Kuroyama-goban	Against	842
do.	9	Kuroyama-yoban	Against	500
do.	10	Kuroyama-samban	Against	1,500
do.	7	Tonaka-niban	Against	1,000(1912)
do.	11	Sambyōme	Against	400
do.	12	Nihyōme	Against	500
do.	16	Dōgo	Both	60
do.	15	Yokobiki-niban	Against	20
do.	13	Ōnakase	Following	248
do.	14	Kaburatobikoe-niban	Following	20
do.	17	Senaka	Following	400
do.	18	Noborihata-kōnoue	Both	1,315
do.	19	Nakashōji	Against	450
Off Fushiki	20	Kojingaeshi	Against	400
do.	21,22	Karimata series	Following	a little
do.	23,24	Suzushima series	Following	a little

The above table shows approximately that the amount caught in the net fixed against the tidal current is double that of the amount caught in that fixed to follow the current. That the latter kind of *Fukube-ami* are fewer in number, is accounted for by the fact that though much liberty is permitted by the authorities, only a few which are profitable in fishing are so fixed, the others bringing so little profit that their fixture would not compensate for the expenditure. These facts seem to show that in this region the former kind of *Fukube-ami* is more efficient in fishing than the latter in this region. This result seems to be caused by the influence of the tidal current, for regard to other external influences, such as the temperature, pressure, food and topographical features of the sea bottom, both kinds of *Fukube-ami* would seem to have equal conditions. In fact, when one observes their movements at

night in the spring, he will see that they are sure to follow the tidal current, for their movements at that time, are not so rapid or active as those of the migratory fish or other cuttle-fish such as the *Ommastrephes* or *Loligo*. They swim slowly, with the head forward, swinging in front the luminous organs of the ventral arms which lighten their course like a lantern.

Comparing the statistics for the last 5 years, the best class of *Fukube-ami*, as to the amount caught in this region, are the following nets: *Kuroyama* series, *Sambyōme*, *Nihyōme* and *Noborihata-kōnoue*, which are in such a position that the school of *Hotaru-ika* very easily enters the nets, the deepest bed of this region being situated very near in front.

In this region, it seems as if the distance of the net from the sea-shore has no special reference to the catch. Is this caused by the fact that the submarine valley is so broad in area that the schools move at the same rate towards the nets of the offing as towards those near coast?

The matter to be noticed with reference to fresh water is that *Dōgo* and *Yokobiki-niban* being washed by the waters of the Shō river, the Shinshō river and the excurrent river of Lake Hōjōzu, stand in a low class, as to the amount caught, but whether that depends upon the fresh water or not, can not be determined until still further evidence has been found. For on the other hand, at Namerikawa there is the interesting fact that the school of feeble *Hotaru-ika* come to the mouth of a river in the late season (June).

That the *Karimata* series and *Suzushima* series are inferior in amount, depends upon the fact that they are very near the shallow beds of the Himi coast, for the same reason as is verified later.

b) *Yokata coast* (Pl. III, fig. 3). This coast has a submarine valley in front as mentioned before, the central portion of which reaches about 100 fathoms and the 50-F-line also approaches very closely to the shore, and the east boundary wall of the valley shows a very sudden incline. The tidal current runs eastwards as is the case with the before mentioned coast. The *Fukube-ami* fixed there at present (1913) are only 9, all facing towards the current, the catch of which is shown in the following table:

Number showing net in map	Name of net	Catch (1913)
2	Ko-ushi	Kan 10,000
3	Makaridashi	5,000 (but 14,000 in average for the late 3 years)
4	Wakura	5,000
5	Natsunaka	1,500 (5,170 in average for the late 3 years)
6	Maenoami	do.
8	Yarigasaki	7,000
9	Goshanomiya	7,000

The average catches are much greater than those of the before mentioned coast. *Ko-ushi* and *Makaridashi* which are superior to the others, have a model situation, facing towards the tidal current and having the deepest bed of the Yokata valley in front. The reference of the catch to the various conditions of the fishing place, agree in the main with those of the before mentioned coast and show an aspect as to verify the former explanation.

c) *Himi coast* (Pl. III, fig. 1). This is the whole coast westward from Ôta village, which is the shallowest, and of which the sea bed has the most gradual incline along the coasts of Toyama Prefecture. And the 50-F-line is generally off the shore as far as 2 or 3 miles, and only on this coast are there found many Molluscan shells. The shore consists mainly of sand, and the water is not so clear as that of Namerikawa.

As this coast is the original part of Toyama Prefecture where the *Fukube-ami* was used, there are many of these nets fixed there. Drag-nets as well as seines from land are also found there, being used as the ordinary fishing implements of the fishermen.

It is not only that the *Hotaru-ika* has been never caught on this coast and that the *Fukube-ami* are only used there to catch other fish such as sardine-allies, but the fishermen have no knowledge of the *Hotaru-ika*. When I went there in May 1913, I asked the fishermen whether they had sometimes observed on this coast the cuttle-fish which discharged light. They answered that they knew well about a kind which they called *Chôchin-ika* (lantern cuttle-fish). So I went in their boat to the fishing place on a dark night, and observed there the

so-called *Chôchin-ika*, which proved to be not the *Hotaru-ika* but a Sepiolid: *Inioteuthis inioteuthis* (Naef). This was really discharging a faint cobaltish light from a great luminous organ which is situated in the mantle cavity near the ink-bag.

The fact that on this coast, the *Hotaru-ika* is not caught, agrees with the fact that the amount of the whole catch by the *Fukube-ami* in the western part of Shimminato coast is very small, as well as for the fact that it is rarely caught on a shallow coast such as Fushiki, and Ishida of Shimoniikawa-gun. And it agrees also inversely with the fact that *Hotaru-ika* are caught in the valleys or on the inclined walls bounding the valleys.

In view of these facts, though the school of *Hotaru-ika* approaches the shallow coast at evening from its dwelling in the daytime, there is some limit in its migration and it seems that it either can not, or will not, move so far as 2 or 3 miles along a shallow bed such as 30 or 40 fathoms.

2. Namerikawa Region.

This region is the coast from Jintsû river eastward to the eastern part of Uozu, occupying the central portion of the whole coast of Toyama Prefecture. The bed of the coast is very deep, the 100-F-line reaching very near the shore, and the valleys are numerous, especially at Namerikawa-cho, where they are so frequent that the boundary of the valley-like inlets shows a zig-zag line. The catch in this region is also very great especially at Namerikawa-cho, where it reaches generally as much as 70 % of the total catch in Toyama Prefecture. Of course this amount arises from the profusion of *Fukube-ami* used there, but also because the catch of each individual net is greater than that of other regions. And the fishermen at Namerikawa make their living mainly by this fishery.

This region is also divided for convenience's sake into 2 coasts: Namerikawa coast and Uozu coast.

a) *Namerikawa coast* (Pl. III, fig. 4). This coast is bounded by 2 shoals, one of which is off Yuwase and the other off the mouth of the Hayatsuki river. *Fukube-ami* of this coast belong entirely to the kind that has the opening in

both sides of the screen-net, being settled in series on the tops of the banks extending toward the offing. The beach consists only of gravel and the sea water is very clear.

The catch of this coast in the following table is the average for 3 years (1911-1913).

Number showing net in map	Name of net	Catch	Number showing net in map	Name of net	Catch
7	Nakase	Kan 4,000	15	Gosha	Kan 7,000
8	Kesagane	5,000	16	Furudatami	9,000
9	Wakizume	15,000	17	Okinoami	12,000
10	Kumajishi	10,000	18	Nakanoami	20,000
11	Nakamurasaki	11,000	20	Nagisaryokei	14,000
12	Komurasaki	18,000	22	Doko	13,000
13	Koami	18,000	23	Takatsuki- nakanoami	20,000
14	Maenoami	7,000			

The *Fukube-ami* which makes the best catch along this coast is always the nearest one to the beach in every series of the net. This is caused, if the preceding various explanations be true, by the fact that the 100-F-line is very near the beach and the tidal current in deeper water is slackened by the submarine banks, so that the *Hotaru-ika* can easily come near the coast along the quite valley-like depression with clear water. And it seems also to depend upon the fact that this coast is situated in the central part of the whole coast in Toyama Prefecture where *Hotaru-ika* are caught.

b) *Uozu coast* (Pl. III, fig. 5). The eastern part of this coast is very deep, the 50-F-line as well as 100-F-line being very near the shore. In this western part, though the valleys are as numerous as at Namerikawa their depth is not so great as there, and the 100-F-line is very far from the shore. There are in this coast about 10 *Fukube-ami*, 2 of which are fixed at the eastern end of this coast and used to catch other fish. The remaining 8 belong either to the *Fukube-ami* following the current or that with a double-opening.

Number showing net in map	Name of net	Catch	Number showing net in map	Name of net	Catch
1	Kôzu	Kan 12,500	5	Okitakase	Kan 2,100
2	Dôgata	4,000	6	Aogake	1,950
3	Nakajima-atari	1,300	7	Fukaguri	1,800
4	Takase Uchitakase	1,800	8	Kawanakajima	2,500

If the table be compared with the map, it will easily be seen that the catch of *Fukube-ami* of this coast is in direct proportion to the degree of its approach to the 100-F-line. It is to be noticed that the net *Kôzu* with the depressions on both sides, reaches in the amount caught nearly to that of the *Fukube-ami* at Namerikawa.

The Hayatsuki river generally contains a very small amount of water, so that its influence upon the net *Nakajima-atari*, seems to be very small.

3. Ikui Region (Pl. III, fig. 6).

This region is the eastern part extending from Kyoden-mura on the whole coast of Toyama Prefecture. The coast is very deep and the tidal current is very rapid, especially so at the eastern coast from Kurobe river. The fishermen of this coast say that the current coming from Noto Peninsula, in some seasons, runs against the Point of Ikuji and it is divided by the point into 2 currents, one of which goes back toward Ishida-mura, making a whirl-current off Ikuji, and the other goes on its course to Echigo. There are at present (1913) only 4 of the ordinary *Fukube-ami* fixed, these all being situated westward from Ikuji-cho, and from what the fishermen say, the coast eastward from Ikuji-sho is not suitable for fixing the ordinary *Fukube-ami*, for the tidal current is so rapid, that the net is driven sideways. The following table shows the average of the catch for the last 3 years (1911-1913).

Fishing place	Number showing net in map	Name of net	Catch
Off Ikuji	1	Sainokami	Kan 1,500
Off the boundary between Ishida and Kyoden	2	Ôshimanoami	400
Off Kyoden	3	Matsunoatari	800
do.	4	Gakenata	600

Hotaru-ika are caught in small quantities by a hauling seine on the beach at Ikuji-cho in the evenings of the fishing season. They are also caught by the same kind of net in great quantity, often to the extent of 50 Kan at Baichi, Kyoden, as mentioned above.

At Tomari farther east from Ikuji-cho, *Hotaru-ika* are often caught by a kind of *fix-net*. And by the investigation of Shimoniikawa District Office, at Iino-mura a special kind of *Fukube-ami*, called "*Ryono-Fukube-ami*", is used made with 2 bag-nets and 2 openings, which is suitable to the coast, the tidal current being very rapid. It was fixed for the first time in 1913, and caught about 35 Kan every evening in spring and sometimes an even greater quantity.

Again at Miyazaki-mura, which is situated near the boundary line between Etchû Province (Toyama Pref.) and Echigo Province (Niigata Pref.), the *Hotaru-ika* is often caught by a drag-net, though the quantity is small.

In view of these facts, though this region is not so well known among those living in Toyama Prefecture as a fishing place for *Hotaru-ika*, as those of other Prefectures, it is clear that in this region also it is caught to some extent. This, according to the preceding explanations, arises from the facts that the 100-F-line is very near the coast and the water is very clear. And the reason that the catch is much less than that of Namerikawa, depends either upon the facts that the current is much more rapid and the banks fewer here than at Namerikawa, or that a net more suitable to that region, would produce a longer catch.

If the preceding facts of all the regions in Toyama Prefecture be summarised, we reach the conclusion that the most favorable coast for the *Hotaru-ika* fishery is (1) one whose 100-F-line is sufficiently near the beach, (2) where some steep depression in a form valley extends at right angles to the shore, and (3) where the tidal current is not too rapid, but where the water is clear enough.

III. SUMMARY.

In view of all the facts stated above, we arrive at the following conclusions:

i) *Hotaru-ika* is a small cuttle-fish, the mantle-length being about 44-54 mm. in the mature male and about 54-67 mm. in the mature female, showing some sexual dimorphisms.

ii) It is a pelagic species, living generally in the deeper water, i. e. 100 or more fathoms, and it is carnivorous.

iii) The ratio of the number of females to that of males caught on the coast differs according to the season: the males are more numerous than the females in late winter or early spring, while the females, on the contrary, increase in number as the fishing season progresses.

iv) They mate probably in early spring.

v) The spawning season is commonly in late April and the whole of May in Toyama Bay, and is also probably the same, or a little earlier in Sagami Bay.

vi) *Hotaru-ika* which are caught in enormous quantities along the coast of Toyama Prefecture in the fishing season, are mostly females mature and mated, each carrying about 14 spermatophores. They are in the deeper water of 100 or more fathoms in the day time, and in the evening, moving as much as possible in deeper water, approach the shore or shallow sea floor to spawn. But they either can not or will not come more than 2 miles along a shallow floor of more than about 30 fathoms. And as soon as they spawn, they retreat to the deep.

vii) The egg is small and ovoid in shape, floating separately on the sea-surface.

viii) The male dies probably after the fixation of spermatophores in the nape of the female, and the female also dies after she has finished spawning.

I must express my opinion as to the eggs and larvae of an *Aburatsubo*

once mentioned by Mr. T. Nishikawa¹⁾. He collected, in early February 1898, many eggs of the *Aburatsubo* of an earlier Embryological stage²⁾—*Nepioteuthis* stage of *Compsoteuthis Nishikawae* of Pfeffer's³⁾—at Aburatsubo Bay, Misaki. And again, he obtained, in April, 1900, like the above, as well as some developed larvae⁴⁾ (*Compsoteuthis Nishikawae* Pfeffer)⁵⁾—*Compsoteuthis* stage of Chun's⁶⁾—at Enoura, Suruga Province. But, judging by the maturation of the specimens from Awa Province and Sagami Bay mentioned before (see p. 89), as they should be mated there in March at the earliest, the first kind of Nishikawa's egg seems to have been collected too early to identify them with the present species *Watasenia scintillans*, while the second kind may be said to be the *Compsoteuthis* stage of the present species for the same reason.

-
- 1). Dôbutsugaku Zasshi, XVIII, 1906, pp. 310-314, pl. 6, figs. 1-15.
 - 2). Ibid. pp. 310-311, pl. 6, figs. 1-11.
 - 3). Erg. Pl.-Exp. Humboldt-Stift. II, F. a, 1912, p. 149.
 - 4). Dôbutsugaku Zasshi, pp. 311-313, pl. 6, figs. 13-15.
 - 5). Erg. Pl.-Exp. Humboldt-Stift. II, F. a, 1912, pp. 450, 162.
 - 6). Wiss. Erg. Deutschen Tiefsee-Exp., Cephal. Oego. 1910, p. 97.

EXPLANATION OF PLATES.

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- Fig. 1. Body color of living female *Hotaru-ika* in spring, nat. size. In the natural specimen it was a little more reddish in color than here painted.
- Fig. 2. Phosphorescence of the same, nat. size.
- Fig. 3. External view of minute organs of mantle of the same, $\times c. 100$.
- Fig. 4. External view of brachial organs of the same, $\times 10$.
- Fig. 5. External view of ocular organs of the same, $\times 2.5$.

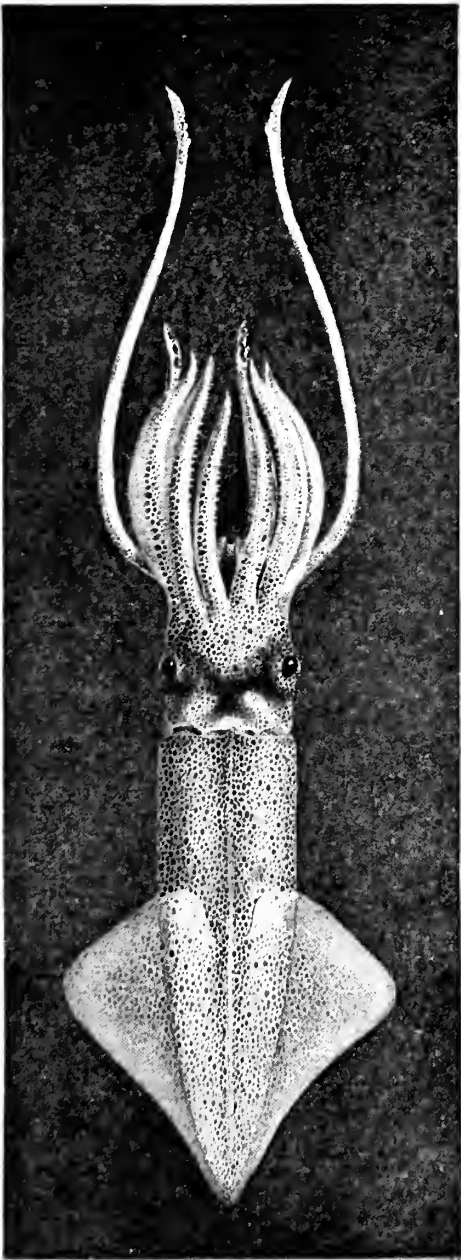
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- Fig. 2. Pen of male, $\times 2$.
- Fig. 3. Hectocotylus, $\times c. 4$.
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- Fig. 11. Inner surface of nidamental gland showing external opening of oviduct.
- Fig. 12. Radula, $\times 80$. Mi) Middle teeth; La) lateral teeth; Mr) marginal teeth.

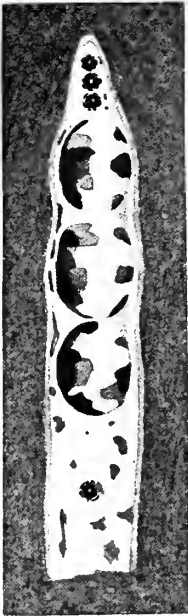
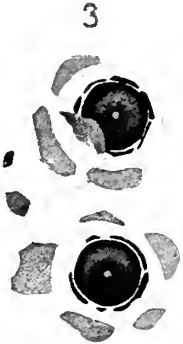
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- Fig. 3. Yokata coast, do.
- Fig. 4. Namerikawa coast, do.
- Fig. 5. Uozu coast, do.
- Fig. 6. Ikuji region, do.





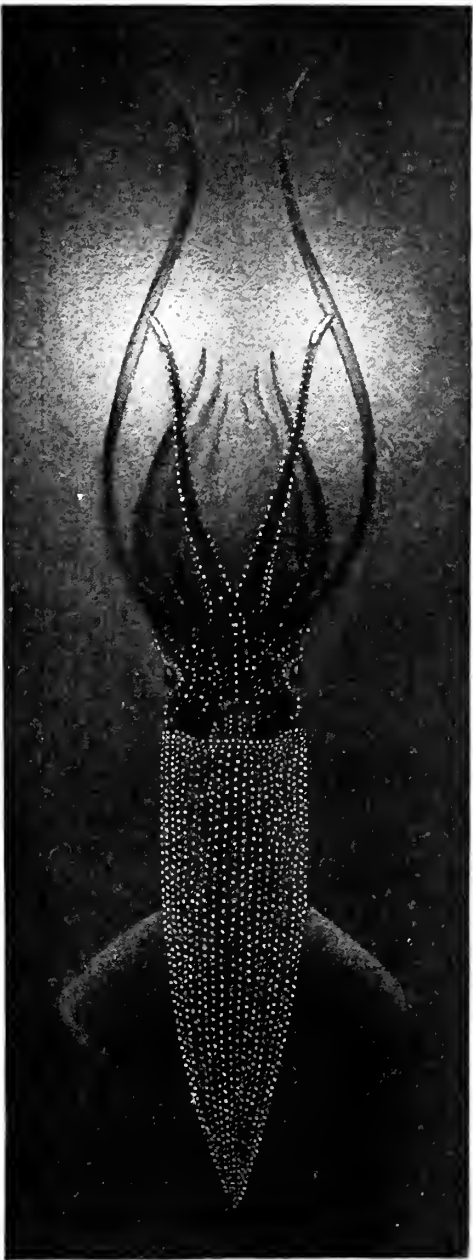
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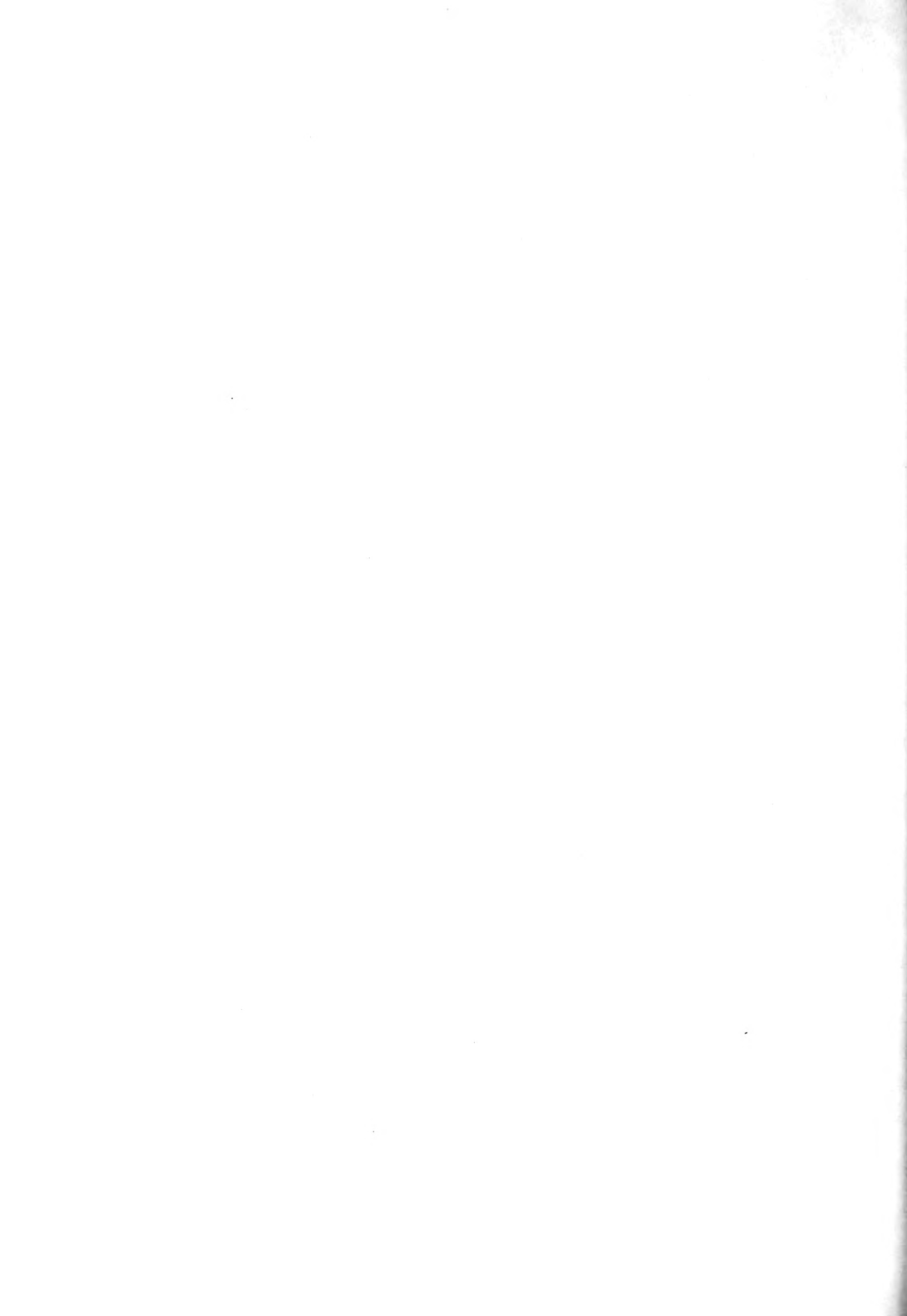
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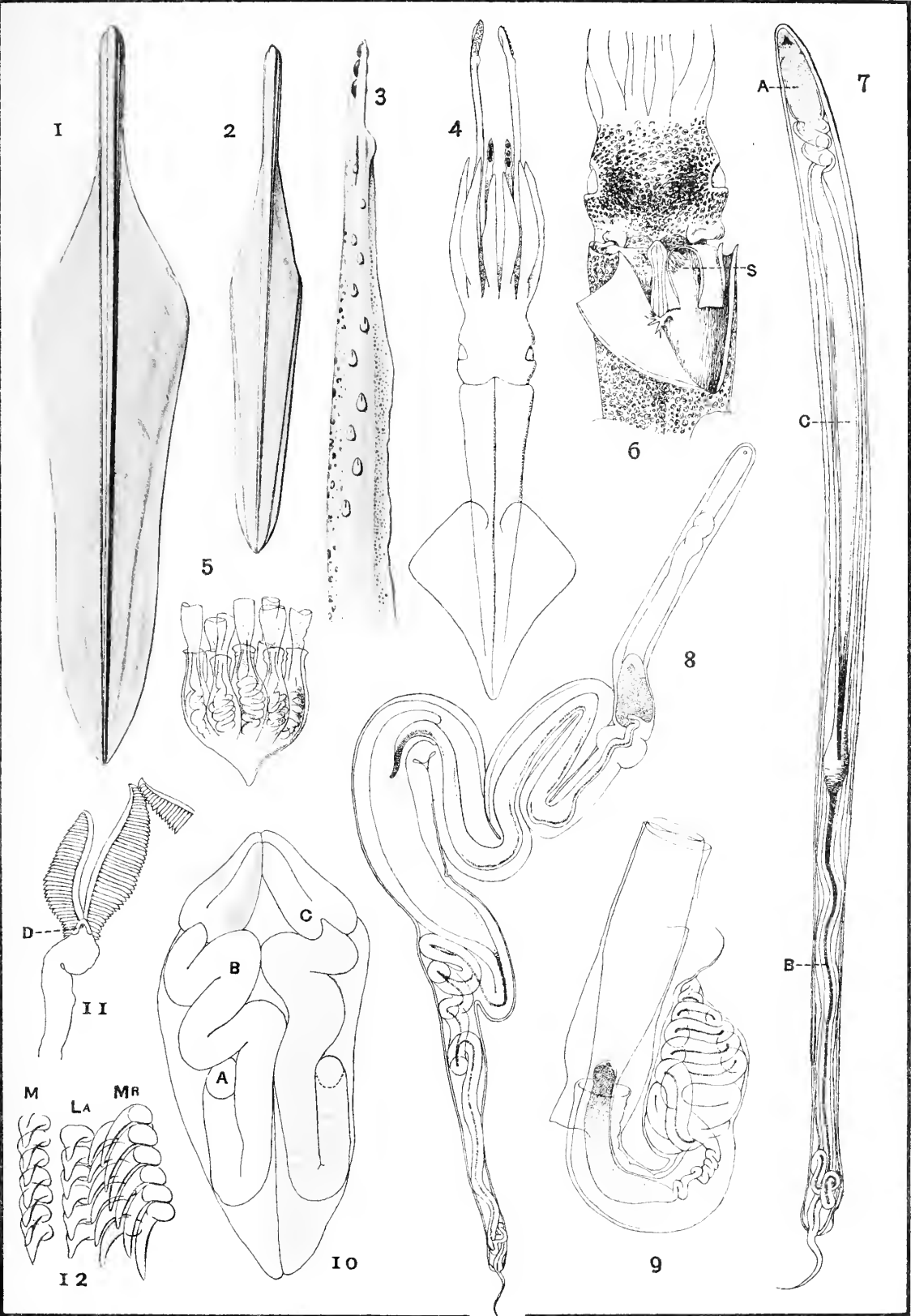


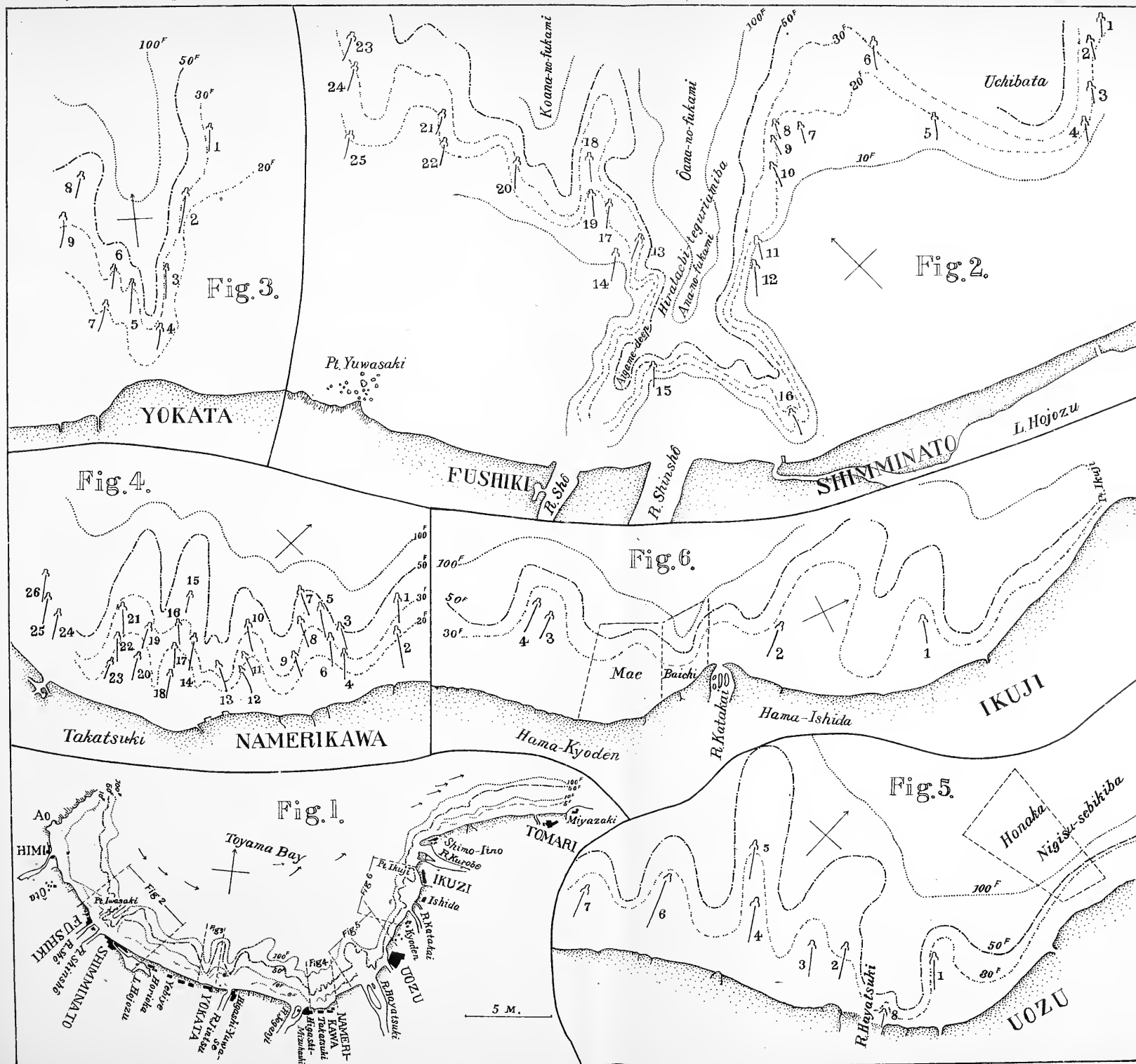
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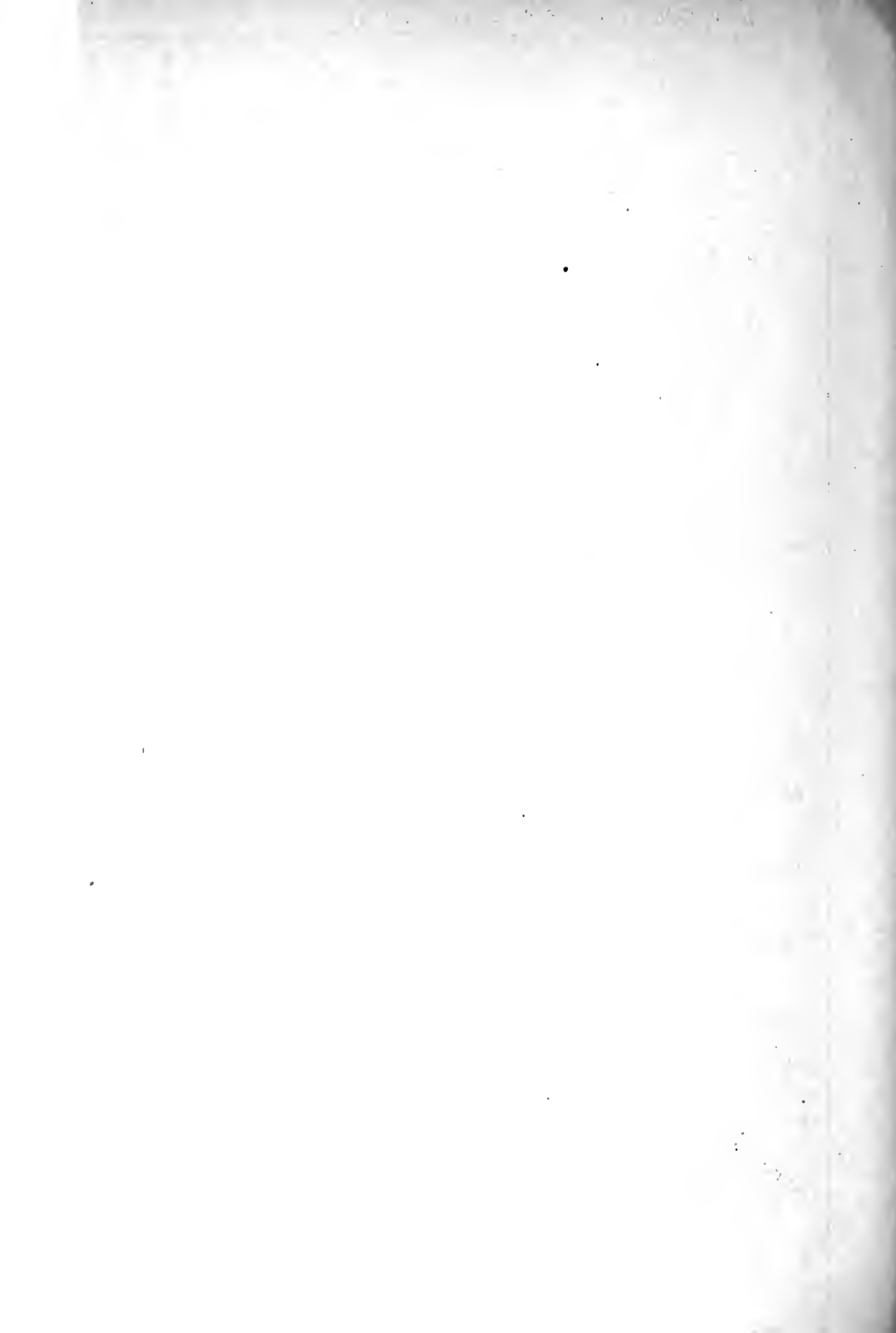


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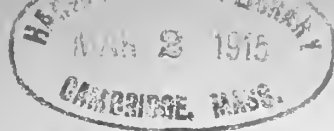
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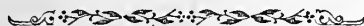


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ÜBER DIE BESTANDTEILE VON FUCUS EVANESCENS

von

Eiji Takahashi, Nōgakushi

Über die chemische Beschaffenheit der Fucus Arten liegen bis jetzt nur einige Untersuchungen vor. Günther und Tollens¹⁾ isolierten zuerst Fucose aus „*Fucus Arten*“, deren Spezies aber nicht angegeben sind. Widtsœ und Tollens²⁾ stellten aus *Fucus serratus* ebenfalls Fucose her, die mit derjenigen identisch ist, die sie im Tragantgummi vorfanden. Keine Forschungen beschäftigen sich mit den in Japan wachsenden Arten.

Im Folgenden veröffentliche ich meine Untersuchungsergebnisse über die Bestandteile von *Fucus evanescens*, welche Tangart in grossen Mengen an der nordöstlichen Küste von Hokkaido: in Kushiro, Nemuro, Kitami und in den Kurilen wächst. Keine Verwertung dieser Art Tang ist bis jetzt in unserem Lande angestrebt worden, obgleich in Europa einige dieser Arten als Düngemittel, Viehfutter und dann besonders auch als Material zur Essigsäuregärung benützt werden. Daher wird es nicht bedeutungslos sein, auch den vorgenannten Seetang in den Kreis der Untersuchung zu ziehen.

Qualitativer Nachweis.

Um einen allgemeinen Begriff zu erhalten, wurde der qualitative Nachweis zuerst durchgeführt.

Abwesenheit von Galaktan wurde durch Oxydation der fettbefreiten

1) Lieb. Ann. **271** (1892) pp. 81-92.

2) Ber. D. chem. Ges. **33** (1900) pp. 132-143.

[Jour. of the College of Agr., Tohoku Imp. Univ., Sapporo, Vol. VI, Pt. 5, December, 1914.]

Substanz mit Salpetersäure von 1,15 spezifischem Gewicht auf gewöhnliche Weise festgestellt.

Etwa 5 gr der Probe wurden mit Aether von Fett und Farbstoffen befreit, mit absolutem Alkohol 30 Minuten lang im Wasserbade gekocht und dann filtriert. Sobald das Filtrat erkaltet war, traten weisse, wolkenartige Krystalle auf, welche nach 24 Stunden abfiltriert und mit Aether ausgewaschen wurden. Der Rückstand bestand aus feinen, seidenglänzenden Nadeln, zeigte sehr süssen Geschmack, schmolz bei 164–165°C und drehte als Lösung die Polarisationssebene nach rechts um. Durch diese Daten ist die Anwesenheit von Mannit deutlich nachgewiesen.

Eine kleine Menge der Substanz wurde in einem Probierrohr zusammen mit verdünnter Salzsäure etwa 10 Minuten lang erhitzt und dann filtriert. Das Filtrat ergab beim Erhitzen mit Phloroglucin und Salzsäure die charakteristischen Reaktionen der Pentose. Die beim Destillieren der Substanz mit Salzsäure von 1,06 spec. Gewicht erhaltene Flüssigkeit zeigte mit Essigsäure-Anilin die charakteristische Farbenreaktion von Furfurol. Das Destillat wurde zunächst mit Hülfe der Spektralreaktion von Oshima und Tollens¹⁾ auf die Gegenwart von Methylfurfurol geprüft. Etwa 10 cc des Destillats wurden unter Zusatz von wenig Phloroglucin und dem gleichen Volumen konz. Salzsäure nach 5 Minuten Stehens filtriert. Das klare, gelbrötliche Filtrat zeigte deutlich das charakteristische Absorptionsspektrum von Methylfurfurol. Beide, Pentosan und Methylpentosan, sind in der Substanz vorhanden.

Quantitative Bestimmung.

In der allgemeinen Analyse wurde folgende Zusammensetzung erhalten :

	In 100 Teilen	In 100 Teilen
	lufttrockner Substanz :	wasserfreier Substanz :
Wasser	3,90	—
Asche	23,30	24,24

1) Ber. D. chem. Ges. **33** (1900) p. 143.

Protein	12,30	12,88
Fett	2,63	2,74
Roh-Faser	4,71	4,90
Stickstoffreier Extraktstoff	53,09	55,24
Gesamtstickstoff	1,93	2,06
Nicht-Eiweisstickstoff	0,41	0,43
Mannit	3,80	3,95
Zellulose	2,76	2,88
Galaktan	Kein	Kein
Pentosan	4,20	4,39
Methylpentosan	4,93	5,13

Das Königsche Verfahren¹⁾ wurde zur Bestimmung der Zellulose angewandt. Demgemäss wurde die Substanz zuerst mit schwefelsäurehaltigem Glyzerin, darauf mit Wasserstoffsuperoxyd, sowie mit Ammoniak behandelt. Pentosan und Methylpentosan wurden nach der Methode von Ellet und Tollens²⁾ bestimmt. Die Substanz wurde mit Salzsäure von 1,06 spec. Gewicht versetzt, bis das Destillat keine charakteristischen Reaktionen von Furfurol und Methylfurfurol mehr gab. Sodann wurde Salzsäurelösung von Phloroglucin dem Destillat zugesetzt, um alles Furfurol und Methylfurfurol als Phloroglucid auszufällen. Am folgenden Tag wurde das Gemisch von Phlorogluciden filtriert, ausgetrocknet und gewogen. Darauf wurde das Methylfurfurol-Phloroglucid vom Furfurol-Phloroglucid getrennt durch Extrahieren des Niederschlags mit 95 % igem Alkohol bei ca 60°C. Für die Berechnung der Pentosanmenge aus der Menge Furfurol-Phloroglucid und der Methylpentosanmenge aus der Menge Methylfurfurol-Phloroglucid wurden die Kröberschen bzw. Ellet und Tollenschen Formeln angewandt.

Mannit wurde nach der folgenden Weise bestimmt: 5 gr der fettfreien Substanz wurden 30 Minuten lang in der Flasche mit 100 cc absolutem

1) Zs. Unters. Nahrungsmittel. Berlin, **12** (1906) pp. 386-395.

2) Ber. D. chem. Ges. **38** (1906) pp. 492-499.

Alkohol ausgekocht, filtriert und wieder mit 100 cc Alkohol versetzt und erhitzt. Die Filtrate wurden gemischt, zur Trockne verdampft und in wenig Wasser gelöst, worauf verschiedene Farbstoffe durch Filtrieren entfernt wurden. Nach der Entfernung von allem Farbstoff durch mehrmalige Behandlung nach der obigen Weise wurde der Rückstand ausgetrocknet und gewogen, hierauf die Menge Asche daraus reduziert.

Wie man oben erschen kann, ist im Vergleich mit andern Seetangarten die Menge an Methylpentosan sehr gross. Zum Vergleiche seien hier auch die Analysen von *Laminaria japonica*, *Kjellmanniella gyrata*, Nori und *Fucus sp.* beigegeben.

	Pentosan	Methylpentosan	Autor
<i>Laminaria japonica</i>	6,42	1,88	S u z u k i ¹⁾
<i>Kjellmanniella gyrata</i>	6,19	2,19	„ ¹⁾
Nori (<i>Pophyra laciniata</i>)	2,59	1,22	Ellet u. Tollens ²⁾
<i>Fucus sp.</i>	6,33	3,46	„ ²⁾
<i>Fucus evanescens</i>	4,37	5,13	T a k a h a s h i

Die Produkte der Hydrolyse.

1) DIE METHODE DER HYDROLYSE.

200 gr des lufttrockenen Seetanges wurden zunächst von mechanischen Verunreinigungen möglichst befreit und darauf mit 2 Liter 4% iger Salzsäure übergossen. Nach 2 Tagen wurde die Flüssigkeit abgegossen und der Seetang so lange mit Wasser gewaschen, bis die abfliessende Flüssigkeit frei von Salzsäure war. Der so gereinigte Seetang wurde sodann mit 4% iger Schwefelsäure 12 Stunden lang in einem Porzellantopf im Wasserbade erhitzt. Der Seetang war nun weich und zeigte den charakteristischen Geruch von Furfurol. Die ganze Masse wurde darauf abgepresst, die so erhaltene hellgelbe Flüssigkeit auf dem Wasserbade etwas erhitzt, mit gefällttem kohlensaurem Kalk neutralisiert und der gebildete Gyps abfiltriert. Dann wurde

1) Trans. Sapporo Nat. Hist. Soc. **1** (1906) p. 122.

2) Ber. D. chem. Ges. **38** (1905) p. 498.

die Flüssigkeit auf dem Wasserbade bis auf ca 200 cc eingengt. Diese Flüssigkeit wurde mit 500 cc 85 % igem Alkohol versetzt, wodurch gummiartige Massen abgeschieden wurden. Nach 24 Stunden wurde die gelblich gefärbte obere Flüssigkeit dekantiert, im Vacuum auf 100 cc eingedampft und der Rückstand wieder mit 500 cc 95 % igem Alkohol vermischt. Eine grosse Menge von braunem Niederschlag trat auf, der nach 12 Stunden langem Stehen abfiltriert wurde. Das Filtrat wurde nun auf 30 cc eingengt und mit 200 cc absolutem Alkohol behandelt. Diesmal schied sich nur wenig Gummi aus, darauf wurde die Flüssigkeit abfiltriert. Das Filtrat wurde sodann weiter bis auf 20 cc eingengt. Der resultierende Syrup war vollständig klar, schwachrot und besass einen salzig-süssen Geschmack,

2) QUALITATIVER NACHWEIS DES SYRUPS.

Der Syrup zeigte die folgenden Reaktionen:

- a) Er reduzierte die Fehlingsche Lösung sehr stark.
- b) Er drehte die Polarisationssebene nach links hin.
- c) Er gab mit Phloroglucin und Salzsäure die charakteristische Farbenreaktion und das Absorptions-Spektrum von Pentose.
- d) Ein Teil des Syrups wurde mit Salzsäure von 1,06 spec. Gewicht destilliert. Das Destillat zeigte mit Phloroglucin und konz. Salzsäure sehr stark die charakteristische Spektralreaktion von Methylfurfurol.
- e) Bei der Oxydation mit Salpetersäure von 1,15 spec. Gewicht trat weder Schleimsäure noch Zuckersäure auf.
- f) Es ergab sich keine Ketose-Reaktion mit Resorcin und Salzsäure.
- g) Er produzierte nicht die charakteristischen Krystalle von Cadmiumbromoxylonat beim Bertrandischen Nachweis ¹⁾.
- h) Kleine Portionen des Syrups wurden auf einem mikroskopischen Objektträger je mit Xylose, Arabinose, Galaktose, Mannose, Glucose und Fucose geimpft. Nach 2 tägigem Stehen sahen die Präparate, mit Ausnahme des mit Fucose geimpften, unverändert aus, im letztern war eine reichliche Ausschei-

1) Bull. Soc. chim. (3) T. 5, p. 546, 554.

dung von Krystallen zu erkennen. Aus den obigen Reaktionen erfolgt, dass der Syrup keine Glucose, Fructose und Galaktose enthielt; dass dagegen das Vorhandensein von Fucose sehr wahrscheinlich ist.

i) Um die Anwesenheit von Fucose ferner zu erweisen, wurde die Hydrazon-Prüfung angewandt. 5 gr des Syrups wurden mit 3 gr Wasser und 3 gr Phenylhydrazin vermischt. Nach 10 Minuten zeigten sich die ersten Krystalle; nach 30 Minuten war das Gemisch fest. Der Niederschlag wurde abgezogen, mit einer geringen Menge von 75 % igem Alkohol ausgewaschen und aus heissem 95 % igem Alkohol umkrystallisiert. Das erhaltene Hydrazon zeigte den Schmelzpunkt bei 172–173°C, während das Fucose-Hydrazon nach der Angabe von Günther und Tollens¹⁾ bei 170–172°C schmilzt. Dadurch erscheint die Gegenwart von Fucose in dem Syrup höchst wahrscheinlich.

3) ISOLIERUNG VON FUCOSE.

Um eine grössere Quantität des Hydrazons zu gewinnen, wurden 2 Kilo des gereinigten Seetanges mit 5 Liter 4 % iger Schwefelsäure nach der oben beschriebenen Weise hydrolysiert und dabei ca 80 cc gereinigten Syrups erhalten.

Nach 2 Wochen wog der Syrup 60 gr. Er wurde nun mit 30 gr Wasser und 30 gr reinem Phenylhydrazin gemischt. Die Flüssigkeit wurde rasch trübe und erstarrte nach Verlauf von 2 Stunden zu einer hellbraun gefärbten Masse. Das Hydrazon wurde mit Aether-Alkohol (3 : 1) zerrührt, abgesogen und mit Aether-Alkohol ausgewaschen, dann nochmals aus 95 % igem Alkohol umkrystallisiert und im Vacuum getrocknet. Der Schmelzpunkt lag bei 171–172°C; das Gewicht betrug 25 gr. Das gereinigte Hydrazon wurde, wie gewöhnlich, mittelst Benzaldehyd zersetzt. Es wurde mit 20 gr Wasser, 38 gr Alkohol und 25 gr Benzaldehyd gemischt und das Gemisch eine Stunde im Wasserbade mit Rückflusskühler erhitzt. Das Hydrazon löste sich schnell beim Erwärmen; beim Abkühlen schieden sich nachher lange Nadeln von Benzaldehydphenylhydrazon vom Schmelzpunkt 155–156°C aus, was die Zersetzung des ursprünglichen Hydrazons bewies. Das Filtrat von diesem

1) Lieb. Ann. **271** (1892) pp. 81–92.

Niederschlag wurde viermal mit Aether ausgeschüttelt und die wässrige Lösung mit Tierkohle gereinigt; sie wurde dadurch ganz farblos, zeigte einen süßlichen Geschmack und reduzierte Fehlingsche Lösung stark. Die Flüssigkeit wurde sodann zu Syrup eingedampft und über Schwefelsäure gesetzt.

Nach 3 Tagen war der Syrup zu einer weissen, krystallinischen Masse erstarrt, die 12,8 gr wog. Diese wurde mit 70% igem Alkohol gemischt und auf einen Tonteller gestrichen. Diesmal sah der Rückstand vollständig rein aus. Darauf wurde dieser in heissem Alkohol gelöst und der Krystallisation überlassen. Schöne weisse, mikroskopisch kleine Krystallnadeln (Gesamtwicht 5 gr) wurden erhalten. Die wurden 2 Wochen über Schwefelsäure getrocknet und im Halbschattenapparat von Schmidt und Haensch polarisiert.

1 gr Substanz wurde in 25 cc Wasser gelöst und in einem 100 mm Rohr polarisiert. Bedeutende Multirotation war vorhanden, nach 24 Stunden betrug die Drehung 8,54 Skalenteile nach links.

$$[\alpha]_D = \frac{-8.54 \times 0.346 \times 25}{1 \times 1} = -73.87^\circ$$

Die spec. Drehung der Fucose ist nach Widtsoe und Tollens¹⁾ fast -74° , der Zucker des Versuches ist folglich nichts anderes als Fucose.

4) NACHWEIS VON ARABINOSE.

Nach der Entfernung der Fucose aus dem Syrup wurde die Mutterlauge auf dem Wasserbade langsam verdampft, bis Alkohol und Aether entfernt waren. Die Lösung wurde mit Wasser vermischt, mit Aether mehrmals ausgeschüttelt, wodurch überschüssiges Phenylhydrazin fast ausgeschlossen war. Die wässrige Flüssigkeit wurde auf dem Wasserbade zu einer dicken, syrupartigen Masse eingengt, welche bei Zusatz von Wasser einen Niederschlag verursachte. Der wurde abgesogen, mit Aether gewaschen und aus 85% igem Alkohol in mikroskopische Nadeln umkrystallisiert. Diese waren blassgelblich gefärbt und schmolzen bei $151-154^\circ\text{C}$. 1,7 gr des Niederschlags wurden mit 1 gr Wasser, 3 gr Alkohol und 1,5 gr Benzaldehyd gemischt und nach der

1) Ber. D. chem. Ges. **33** (1900) p. 141.

oben beschriebenen Weise zersetzt. Die Zersetzung gelang gut. Die von Benzaldehydphenylhydrazon abfiltrierte Lösung wurde dreimal mit Aether ausgeschüttelt und nach Reinigung mit Blutkohle sorgfältig zu Syrup eingedampft.

Um Sicherheit zu haben, dass der resultierende Syrup Pentose enthielt, wurde ein Tropfen desselben der Pentose-Probe mit Phloroglucin und Salzsäure unterworfen, wobei durch die charakteristische Farbenreaktion und die Absorptionsstreifen im Spektrum das Vorhandensein von Pentose-Gruppen nachgewiesen wurde. Sodann wurden zwei Tropfen des Syrups auf einen mikroskopischen Objektträger je mit einem Kryställchen von Xylose und Arabinose geimpft. Nach 3 Tagen war der mit Xylose geimpfte Tropfen noch unverändert, der mit Arabinose behandelte dagegen enthielt eine reichliche Ausscheidung von nadelförmigen Krystallen, die jeden Tag ausgedehnter wurde.

Aus diesem Versuche geht hervor, dass das Vorhandensein von Arabinose in dem Syrup wahrscheinlich ist, doch konnte ich weitere Versuche der zu geringen Menge Syrup wegen leider nicht vornehmen.

Zusammenfassung der Ergebnisse.

Fassen wir nun die Resultate der Untersuchungen zusammen, so ergeben sich folgende Grundtatsachen:

- 1) Da Pentose und Methylpentose in den Produkten der Hydrolyse von *Fucus evanescens* nachgewiesen wurden, ist die Anwesenheit von Pentosan und Methylpentosan deutlich.
- 2) Da eine ziemlich grosse Menge von Fucose aus dem Syrup isoliert wurde, besteht das Methylpentosan vorzüglich aus Fucosan.
- 3) Arabinose wurde auch nachgewiesen, was die Anwesenheit von Arabin wahrscheinlich macht.
- 4) Mannit ist vorhanden in dem Seetang, aber kein Galactan.

Zum Schluss möchte ich Herrn Prof. Dr. K. Oshima meinen besten Dank aussprechen für seine liebenswürdige Unterstützung bei der Ausführung dieser Untersuchungen.

ÜBER DIE KOLLOIDALEN EIGENSCHAFTEN DER SAUREN BÖDEN IN JAPAN

Mitteilung II

von

Tetsutaro Tadokoro, *Nōgakushū*

In meiner vorigen Arbeit ¹⁾ habe ich über die kolloidalen Eigenschaften der mineralischen sauren Böden in Japan Bericht erstattet. Die nämlichen Versuche wurden an anderen sauren Bodenproben, die aus Taiwan (Formosa) stammen, ausgeführt. Zur Untersuchung gelangten gleicherweise auch 3 Proben neutralen oder schwach sauren Bodens.

Die Resultate, die sich bei der Untersuchung der 14 Proben saurer Böden ergaben, wurden mit denjenigen der 3 neutralen Böden verglichen.

Die vorliegende Arbeit soll über die Verschiedenheiten zwischen den kolloidalen Eigenschaften der sauren Böden einerseits und den neutralen Böden andererseits Auskunft geben. Über die kolloidalen Lösungen gedenke ich in nächster Zeit ausführlichen Bericht zu erstatten.

Wahl der Proben.

Bei der Auswahl des Untersuchungsmaterials wurden drei Proben von neutralen Böden, ausserdem zwei mineralische stark saure Böden aus Formosa ausgewählt. Die erste Probe bestand aus Sand-, die zweite aus Ton- und die dritte aus Humusboden. Jede frische Probe wurde an der Luft vorgetrocknet und durchgesiebt. (Dia. 0,5 mm.)

1) Jour. of the College of Agr. Tohoku Imp. Univ., Sapporo, Vol. VI, Pt. 2, June, 1914.

[Jour. of the College of Agr., Tohoku Imp. Univ., Sapporo, Vol. VI, Pt 5, December, 1914]

Proben	Wassergehalt %	Azidität n. d. Daikuhara-KCl- methode ccm	Sammelort	採 集 地
13. Sandboden	3,54	0,49	Hokkaido, Sapporo.	北海道札幌區東北帝國 大學農科大學第一農場
14. Tonboden	5,22	2,08	„ „	„
15. Humusboden	5,43	4,83	„ „	„
16. saurer Boden (Laterit)	3,69	104,92	Taiwan, Taihoku, Takarappo.	臺灣 臺北廳 大加納 堡 後山庄
17. saurer Boden (Laterit)	3,01	47,90	Taiwan, Toenchō, Chikuhoku- niho.	臺灣 桃園廳 竹北二 保 楊梅厝庄

Versuch 1. Quellung.

Zur Prüfung des Quellungsvermögens dieser Proben wurde, wie im vorigen Berichte erwähnt ist, ein Gemisch von 5 g des Bodens mit 15 ccm des Reagens 30 Minuten lang geschüttelt, worauf nach 24 Stunden das vergrößerte Volumen bestimmt wurde.

Tabelle 1.

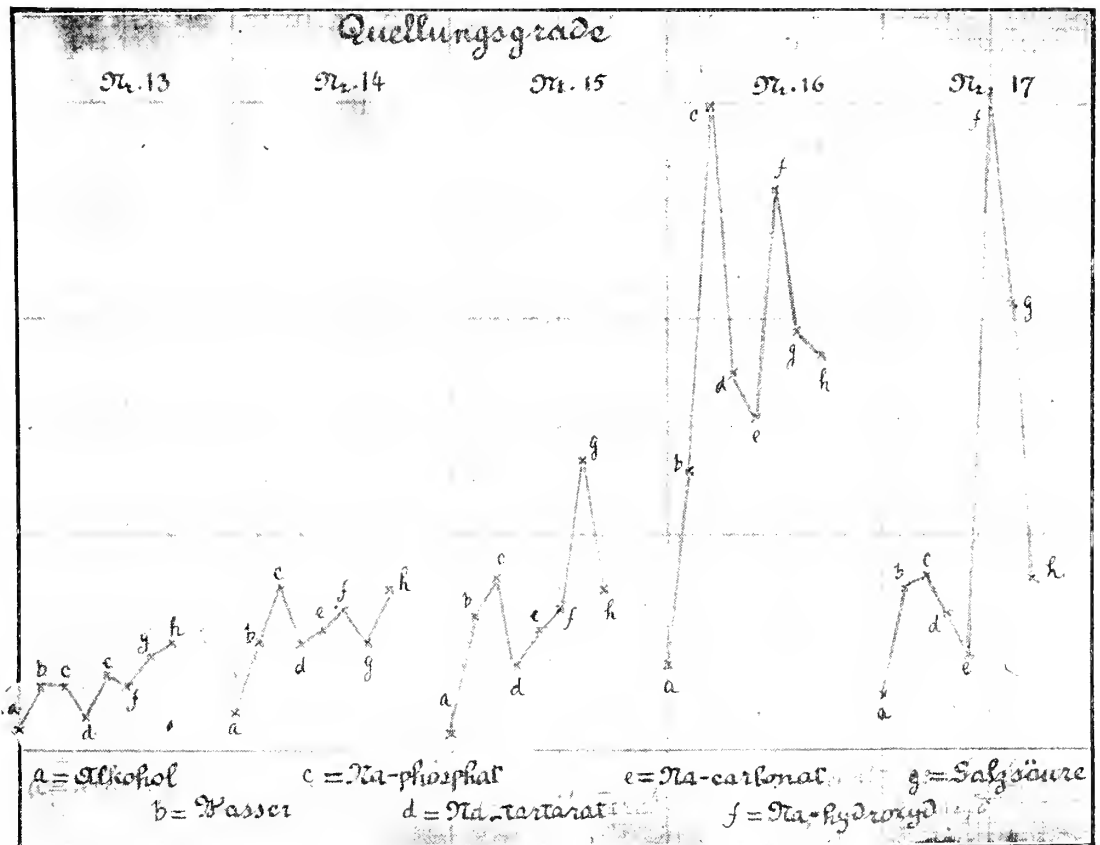
Nummern der Proben	Quellung mit Alkohol	Quellung mit Wasser	mit 5 % Na Phos- phat-lg.	mit 5 % Na-Ac- etat-lg.	mit 5 % Na-Tar- tat-lg.	mit 5 % Na-Car- bo. nat-lg.	mit 5 % NaOH	mit 5 % HCl
13.	5,1	5,5	5,5	5,2	5,6	5,5	5,8	5,9
14.	5,8	6,5	7,0	6,5	6,6	6,8	6,5	7,1
15.	6,1	7,4	7,8	6,8	7,2	7,5	9,0	7,7
16.	5,6	7,3	10,6	8,2	7,8	9,8	8,6	8,4
17.	5,2	6,1	6,2	5,9	5,5	10,0	8,6	6,2
Prozentzahlen der vergrößerten Volumen der geglihten Proben :								
13.	4,0	12,2	12,2	6,1	14,0	12,2	18,3	20,4
14.	7,4	20,3	29,6	20,3	22,2	25,9	20,3	31,1
15.	3,4	25,4	32,2	15,9	22,0	27,1	51,2	30,5
16.	16,6	52,0	120,8	70,8	62,5	104,1	78,3	74,1
17.	10,8	10,8	31,9	25,5	17,0	122,7	82,9	31,8

Die obigen Werte, sowie die vergrößerten Volumen, wurden in den

Vergleichungen aus folgenden gegliihten Proben berechnet.

Nummern der Proben	13.	14.	15.	16.	17.
Quellungsvolumen der gegliihten Proben	4.9	5.4	5.9	4.8	4.7

Wenn in der untenstehenden Figur die vergrösserten Volumen auf der Y Achse und die verschiedenen Reagentien auf der X Achse aufgetragen werden, so kann man aus jeder Kurve aus ihrer Entfernung von der X Achse die Werte leicht erschen.



Zum Nachweis wollen wir die freien Humussäuremengen angeben; die folgenden Werte sind die Prozentzahlen der trocknen Substanzen.

Nummern der Proben	13.	14.	15.	16.	17.
% der freien Humussäuremengen	0,57	2,37	5,52	2,12	2,32

Versuch 2. Hygroskopizität.

Um die Beziehungen der Hygroskopizität zum Kolloidgehalt zu erkennen, unternahmen wir folgenden Versuch, wobei die Bestimmung der Hygroskopizität nach dem Verfahren von H. Rodewald und A. Mitscherlich erfolgte.

Das Verfahren besteht in folgenden Grundzügen: Es wird zunächst der lufttrockne Boden in einem mit Phosphatpentoxyl versehenen Exsikator behandelt und dann in den 10 0/0 ige Schwefelsäure enthaltenden Apparat gestellt und die aufgenommene Wassermenge bestimmt.

(Vergl. J. König: Unters. landw. gewerb. Stoffe. Vierte Aufl. S. 76)

Nummern der Proben	13.	14.	15.	16.	17.
Gehalt in Prozenten	4,99	7,84	6,77	6,33	4,48

Aus diesen Zahlen ergibt sich, dass die Hygroskopizität in der Probe 2 am höchsten und in der Probe 5 am geringsten ist. Die Durchschnittszahl beträgt 6,08 0/0.

Tabelle 2.

Vergleichung der Quellungsgrade mit der Hygroskopizität.

Nummern der Proben	Hygroskopizität = HY	Quellungsvolumen = QV	Verhältnisse = $\frac{QV}{HY}$
13.	4,99	5,5	1,11
14.	7,84	6,5	0,83
15.	6,77	7,4	1,09
16.	6,33	7,3	1,15
17.	4,48	6,1	1,14

Diese Vergleichen zeigen, dass die Verhältnisse einander sehr nahe kommen. Sie lassen ferner erkennen, dass die Hygroskopizität gewöhnlich in nahen Beziehungen zum Quellungsgrade steht.

Versuch 3. Adsorption.

Zur Bestimmung der Adsorptionsfähigkeit des Kolloides für Farbstoffe hat der Verfasser Eosin als Färbemittel ausgewählt. Die Bestimmung der Farbstoffkonzentration kann zweckmässig auf kolorimetrischem Wege durch Vergleich mit den Kontrollösungen ausgeführt werden. Für die Farbstofflösungen werden zwei Konzentrationen, nämlich zu 1,0 und 2,0 g im Liter, verwendet. Wenn auch die Temperatur und die Berührungszeit bei der Adsorption keine so grosse Rolle spielen wie bei anderen physikalischen und chemischen Vorgängen, so werden wir doch bei konstanter Temperatur arbeiten und die Farbstoffe stets zu gleicher Zeit einwirken lassen. Das Gemisch von 5 g Boden und 20 ccm Farbstofflösung wird jedesmal ca eine halbe Stunde geschüttelt, 6 Stunden stehen gelassen und bei Zimmertemperatur (15-17°C) geprüft.

Tabelle 3.

Nummern der Proben	Bei unadsorbiert gebliebenen Farbstoffmenge entsprechend, 1/2 % und 1 % Farbstofflösung in 5 ccm Wasser zurücktitriert. Die Werte bezeichnen die Mengen der Farbstofflösungen.	
	1/2 % ige Farbstofflösung	1 % ige Farbstofflösung
13.	2,8	5,0
14.	2,2	7,0
15.	1,8	6,0
16.	1,3	3,2
17.	1,6	4,5

Für die Herstellung des Adsorptionsgleichgewichtes kommen wir dann empirisch zu der Gleichung :

$$X/M = K \cdot C^{1/n} \quad ; \quad K = \frac{X/M \text{ (adsorbiert)}}{C^{1/n} \text{ (frei)}}$$

In welcher Gleichung X die Farbstoffmenge, die von 1 mg Boden aus einer Lösung adsorbiert wird und C die Konzentration (mg in 1 ccm) des

Farbstoffes im Wasser nach der Adsorption und $1/n$ der Exponent (Vergl. Versuch 4) ist. Für die rechnerische Darstellung der Adsorptionsvorgänge wird obige Gleichung logarithmiert. So erhalten wir

$$\log K = \log X/M - 1/n \log C$$

Tabelle 4.

Die folgenden Werte wurden gefunden :

Nummern der Proben	1/2 % ige Farbstofflösung		1 % ige Farbstofflösung.	
	X/M	C	X/M	C
13.	2,11/5000	5,38	3,75/5000	7,50
14.	2,93/5000	3,05	6,25/5000	5,84
15.	3,54/5000	2,64	6,83/5000	5,45
16.	4,41/5000	2,06	8,15/5000	3,90
17.	3,87/5000	2,42	7,05/5000	4,73

Tabelle 5.

Vergleichung von Quellungsvolumen und Hygroskopizität mit der Adsorption von Farbstoff.

Nummern der Proben	X, M · 1000	Quellungs- volumen	Hygroskopizi- tät	Verhältnisse	
				(X/M·1000) : (QV)	(X/M·1000) : (HY)
13.	0,75	5,5	4,99	0,136	0,150
14.	1,05	6,5	7,84	0,161	0,135
15.	1,36	7,4	6,77	0,183	0,200
16.	1,63	7,3	6,33	0,223	0,257
17.	1,41	6,1	4,48	0,231	0,314

Die Verhältnisse zwischen Adsorption und Quellungsvolumen kommen einander in allen Proben sehr nahe ; ebenso die zwischen Adsorption und Hygroskopizität.

Versuch 4. Ammoniakabsorption.

Zur Bestimmung der Ammoniakabsorptionskraft von diesen 5 Proben führten wir den nämlichen Versuch durch, wie er schon im vorigen Bericht beschrieben worden ist.

100 g Erde wurden mit 250 ccm normaler Ammonchloridlösung gemischt, geschüttelt und fünf Tage lang stehen gelassen. Dann wurden 25 ccm verdünnten Extrakts in einen 250 ccm haltigen Messkolben gefüllt. Das Ammoniak wurde bestimmt durch Destillation mit 50 ccm Wasser und frisch gebrannter Magnesia. Das Ammoniak wurde mit 1/10 Normalschwefelsäure aufgefangen und mit 1/10 Normalnatronlauge zurücktitriert.

Tabelle 6.

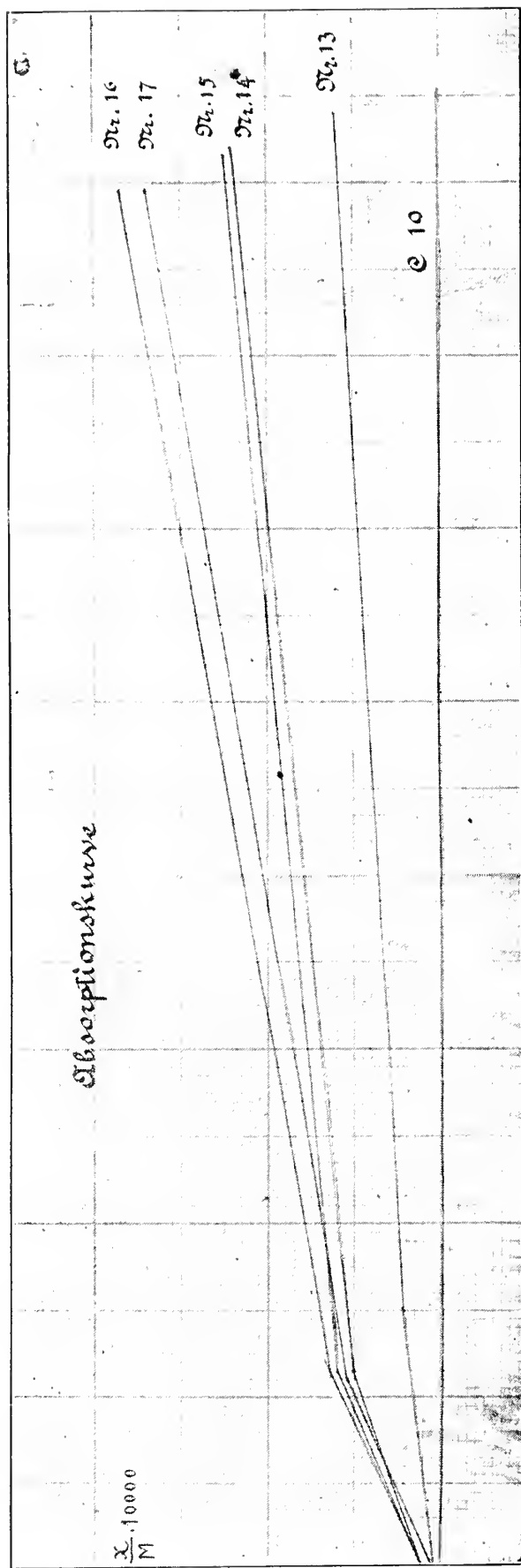
Nummern der Proben	Absorptionskoeffizient		
	in N/100 NH ₄ Cl Lösung	in N/10 NH ₄ Cl Lösung	in Normal NH ₄ Cl Lösung.
13.	6,72	43,97	127,69
14.	19,82	104,13	241,06
15.	20,15	117,56	252,06
16.	20,48	124,27	271,21
17.	19,58	111,84	247,82

Nach Herstellung des Absorptionsgleichgewichts erhalten wir folgende Zahlen : (Vergl. Versuch 3)

Tabelle 7.

Nummern der Proben	(I) N/100 NH ₄ Cl Lösung		(II) N/10 NH ₄ Cl Lösung		(III) Normal NH ₄ Cl Lg.	
	X/M	C	X/M	C	X/M	C
13.	0,000067	0,0735	0,000439	1,5624	0,00127	16,87
14.	0,000198	0,0473	0,001041	1,3180	0,00241	16,42
15.	0,000202	0,0465	0,001176	1,322	0,00252	16,37
16.	0,000205	0,0459	0,001243	1,241	0,00371	15,90
17.	0,000196	0,0473	0,001118	1,291	0,00347	15,95

Wir wollen die graphische Darstellung für diese Vorgänge wählen, dabei ergeben sich die folgenden Daten :



Wie die obige Figur zeigt, nimmt die Nährstoffabsorptionskraft wie die Farbstoffabsorptionskraft mit der Verdünnung der Konzentration ab, der Beziehungswert ist daher in jeder Probe gleich. Aus obigen Angaben wird der Wert von $1/n$ und K nach folgenden Gleichungen berechnet. Die schon erwähnte Gleichung ist:

$$K = \frac{X/M}{C^{1/n}} \quad ; \quad X/M = K \cdot C^{1/n}$$

Zuerst wird der Wert von $1/n$ nach folgender Gleichung berechnet:

$$\frac{X_1}{M} = K \cdot C_1^{1/n} \quad ; \quad \frac{X_2}{M} = K \cdot C_2^{1/n}$$

$$\frac{X_1}{X_2} = \frac{C_1^{1/n}}{C_2^{1/n}} \quad ; \quad \log X_1 - \log X_2 = 1/n (\log C_1 - \log C_2)$$

$$1/n = \frac{\log X_1 - \log X_2}{\log C_1 - \log C_2}$$

Tabelle 8.

Nummern der Proben	$1/n$ =eliminiert aus (I) & (II)	$1/n$ =eliminiert aus (II) & (III)	$1/n$ =Durchschnittszahl
13.	0,2890	0,2786	0,2838
14.	0,3148	0,2171	0,2695
15.	0,3304	0,1480	0,2392
16.	0,3468	0,2607	0,3035
17.	0,3340	0,2819	0,3095

Dann folgt die Berechnung des Wertes von K mit obiger Durchschnittszahl nach folgenden Gleichungen:

$$K = \frac{X/M}{C^{1/n}} \quad ; \quad \log K = \log X/M - 1/n \log C$$

Tabelle 9.

Nummern der Proben	K=eliminiert aus (I)	K=eliminiert aus (II)	K=eliminiert aus (III)
13.	0,000140	0,000097	0,000561
14.	0,000448	0,0009661	0,001133
15.	0,0004201	0,002595	0,001291
Durchschnitts- zahl d. obigen drei Proben	0,0003506	0,001510	0,00099
16.	0,0002611	0,001164	0,001606
17.	0,000388	0,001033	0,001465
Durchschnitts- zahl d. obigen zwei Proben	0,0003245	0,001098	0,001535

Zusammenfassung.

Zur Erkennung der speziellen kolloidalen Eigenschaften der mineralischen sauren Böden müssen wir sie mit verschiedenen neutralen Böden vergleichen.

Wenn man die Resultate des vorigen Berichtes mit denen des vorliegenden vergleicht, so ergibt sich folgende Gesamtübersicht. An erster Stelle wollen wir über die allgemeinen kolloidalen Eigenschaften der japanischen sauren Böden sprechen und sodann die Unterschiede zwischen den kolloidalen Eigenschaften der mineralischen sauren Böden und den verschiedenen neutralen Böden diskutieren.

I DIE ALLGEMEINEN KOLLOIDALEN EIGENSCHAFTEN.

1) Beim Quellungsversuch benützten wir die folgenden 9 Reagentien : Alkohol, Wasser, Na-Phosphat, Na-Tartarat, Na-Carbonat, Natronlauge, Salzsäure und Essigsäure und beobachteten, dass die Quellungsgrade der verschiedenen Böden am grössten sind in Alkalien wie in Natronlauge und in Na-Carbonatlösung, dann absteigend in Säuren, Salzlösungen, Wasser und am kleinsten in Alkohol.

2) Die Resultate der Quellungsversuche zeigen uns, dass die gebildeten Gelformen je nach der Art des Reagens verschieden sind und die Gelbildungsmaterialien des Bodens nicht einheitliche Substanzen darstellen.

3) Die Unterschiede der vergrösserten Volumen zwischen den originalen Proben und den mit Wasser extrahierten Resten sind sehr klein. Daher haben die kolloiden Substanzen, die aus dem Boden mit reinem Wasser extrahiert werden, keine wichtige Wirkung auf die Quellung des Bodens.

4) Die Quellungswärme, die wir von der Benetzungswärme unterscheiden müssen, ist wahrscheinlich proportional dem Quellungsgrade für die verschiedenen Böden.

5) Im Einklang mit J. M. van Bemmels Angabe nimmt die Adsorptionskraft des Bodens für Farbstoffe wie auch für Nährlösungen mit der Verdünnung ihrer Konzentration ab. Die Verhältnisse zwischen Farbstoffadsorption und Quellungsvolumen kommen sich sehr nahe, wie auch die zwischen Farbstoffadsorption und Hygroskopizität. Diese Erscheinungen zeigen uns, dass die Grösse der Quellungsgrade, die Hygroskopizität und die Farbstoffadsorption in inniger Beziehung zur Menge der festen kolloidalen Substanzen des Bodens stehen.

6) a) Die Ammoniakabsorptionskraft des Bodens hat keine regelmässige Beziehung zu den Farbstoffadsorptionen.

b) Die Ammoniakabsorptionskraft des Bodens hat keine regelmässige Beziehung zum Quellungsvolumen und zu der Hygroskopizität.

c) Aus dieser Tatsache erkennen wir, dass die Ammoniakabsorptionserscheinungen kompliziertere Beziehungen zeigen als die Farbstoffadsorption, die nur mit der Oberfläche der Teile im kolloiden Zustande zusammenhängt.

7) Der Ammoniakabsorptionskoeffizient der Böden für Normalammonphosphatlösungen ist grösser als der für Normalammonchloridlösungen. Und es ist wohl anzunehmen, dass bei Ammonphosphatbehandlung die freige-wordene Phosphorsäure sich mit dem Aluminium verbindet und diese unlösliche Verbindung die Ammoniakabsorptionskraft des Bodens vergrössert.

8) Bei neutralen Böden können wir je nach der Bodenart die folgenden Unterschiede in den kolloidalen Eigenschaften erkennen :

a) Die Zunahme des Quellungsvolumens ist am grössten im Humusboden, geringer im Tonboden und am kleinsten im Sandboden.

b) Die Hygroskopizität ist am grössten im Tonboden, dann im Humusboden und am kleinsten im Sandboden.

c) Die Farbstoffadsorptionskraft ist am stärksten im Humusboden, etwas geringer im Tonboden und am schwächsten im Sandboden. (Die Unterschiede zwischen Sandboden und Tonboden sind sehr gross.) Die Zunahme der Farbstoffadsorption nach den Konzentrationsänderungen ist beim Sandboden am kleinsten.

d) Der Ammoniakabsorptionskoeffizient ist beim Humusboden am grössten und im Sandboden am kleinsten.

II SPEZIELLE KOLLOIDALE EIGENSCHAFTEN DER MINERALISCHEN SAUREN BÖDEN.

1) Die Quellungsgrade des mineralischen sauren Bodens sind gewöhnlich grösser als beim humusreichen neutralen Boden, dessen vergrössertes Volumen am höchsten unter allen verschiedenen neutralen Böden ist ; dieser Unterschied ist am grössten in der Behandlung mit Na-Carbonatlösung und mit Natronlauge. Aber andererseits ist das Quellungsvolumen einiger sauren Bodenarten kleiner als das der humusarmen neutralen Böden.

2) Bei mineralischen sauren Böden sind die Unterschiede der Quellungsvolumen je nach den Reagentien viel grösser als bei neutralen Böden. Diese Erscheinungen zeigen uns, dass die mineralischen sauren Böden reichliche Mengen von veränderlichen kolloidalen Substanzen haben d. h. die kolloidalen Substanzen der mineralischen sauren Böden haben bewegliche, unbeständige Formen.

3) Die Hygroskopizität der verschiedenen mineralischen sauren Böden

zeigt keine beträchtlichen Unterschiede verglichen mit den neutralen Böden. Aber die Verhältnisse der Quellungsvolumen zur Hygroskopizität sind bei mineralischen sauren Böden gewöhnlich grösser als bei neutralen Böden, ausser beim humusreichen sauren Boden.

4) Die Adsorptionskraft des mineralischen sauren Bodens für Farbstoffe ist gewöhnlich grösser als beim neutralen Boden, die Adsorptionskraft des humusreichen neutralen Bodens ist der kleinsten Kraft der mineralischen sauren Böden entsprechend.

5) Die Verhältnisse der Adsorptionskraft zur Hygroskopizität berechnet für die mineralischen sauren Böden sind gewöhnlich grösser als beim neutralen Boden, ausser beim humusreichen sauren Boden.

6) Die Ammoniakabsorptionskraft zeigt gewöhnlich grössere Werte beim mineralischen sauren Boden als beim neutralen Boden. Aber zwischen dem kleinsten Werte des erstern und dem grössten Werte des letztern bestehen keine beträchtlichen Unterschiede.

7) Beim humusreichen sauren Boden und beim neutralen Boden ist die Zunahme des Ammoniakabsorptionskoeffizienten nach der Konzentration nicht grösser als beim mineralischen sauren Boden.

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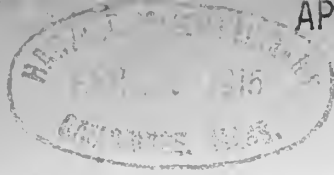
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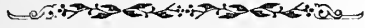


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-

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ON THREE INTERESTING NEW OEGOPSIDS FROM THE BAY OF SAGAMI.

By

Madoka Sasaki, *Rigakushi*.

~~~~~  
With 1 ✓ plate and 4 text-figures.

In the collection of cuttle-fishes preserved in the Museum of the Science College, Tokyo Imperial University, three interesting species of Oegopsids caught in Sagami Bay are found, which seem to be as yet unclassified, as shown by the following description.

## ***Meleagroteuthis separata*** sp. nov.

*Type*.—A single male specimen obtained by Mr. KUMAKICHI AOKI from a depth of about 400 fathoms at Uchibata, Misaki, March 4, 1898.

The measurements are as follows:

|                                |       |                  |
|--------------------------------|-------|------------------|
|                                | mm.   |                  |
| Dorsal length of mantle.....   | 31    |                  |
| Ventral length of mantle ..... | 25    |                  |
| Breadth of mantle .....        | 20    |                  |
| Ventral length of head .....   | 19    |                  |
| Breadth of head .....          | 22    |                  |
| Total breadth of fins .....    | 24    |                  |
| Length of fins.....            | 14    |                  |
| Length of first arm .....      | 52    | left right<br>50 |
| „ „ second arm .....           | 52(?) | 49 (?)           |
| „ „ third arm .....            | 56    | 52 (?)           |
| „ „ fourth arm.....            | 40    | 47               |
| „ „ tentacle .....             | 80    | 80               |

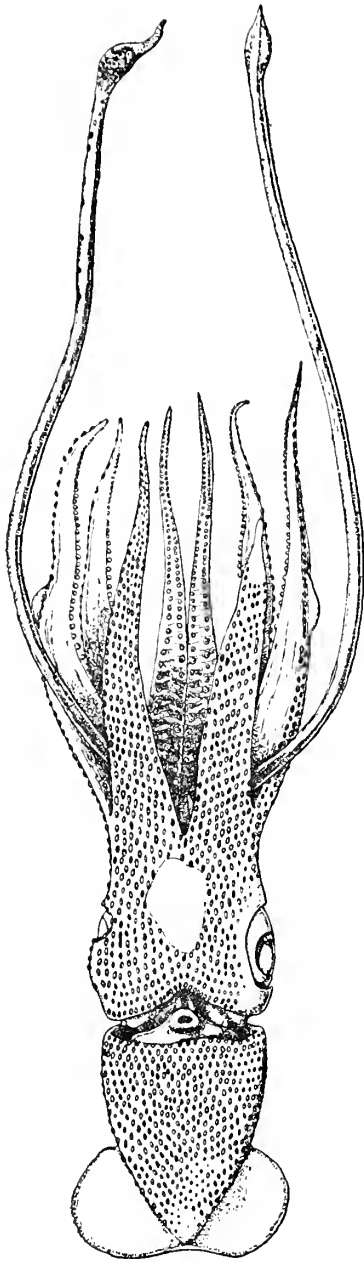


Fig. 1.—*Melagrotenthis separata* sp. nov.  
ventral aspect with luminous organs  
in approximate position.

The mantle is of a conical shape, broadest a little back from the anterior margin, and the length is about one and a half times greater than the breadth; the dorsal anterior margin projects a little into an obtuse angle in the middle. The fins are semicircular, extending a little backwards beyond the posterior end of the mantle, and their length is a little shorter than half the mantle-length (text-fig. 1).

The head is large, and broader than the mantle-opening, and it is clearly marked from the neck by a boundary edge, the ventral part of which shows a low arch bounding a shallow siphonal groove.

The eyes are very asymmetrical, the eye-opening as well as the eye-ball of the left side being twice or more greater than those of the right side in diameter. On each side of the neck there is a low transverse ridge with an olfactory papilla. The siphon is short and conical, connected with the siphonal groove, by two pairs of dorsal ligaments somewhat submedian in position. The siphonal resisting cartilages are nearly oval in shape, being a little acuminate anteriorly, with an elongate deep median depression; the nuchal cartilage is lanceolate, a little tapering posteriorly. The mantle cartilages corresponding with the siphonal ones are high and linear

but short, being about as long as the siphonal ones.

The arms are subequal and long, the longest arm being about twice as long as the mantle-length, and the order of length seems about  $3 > 2 > 1 > 4$ , though the extremities of some arms are broken in the type specimen. They taper gradually towards the extremities and are nearly quadrangular in section, the outer surfaces being provided with neither webs nor keels; but three dorsal pairs of arms have, along their median lines, a series of horny tubercles, details of which will be given subsequently. The third pair of arms has a short semilunar web in the middle of the outer surface.

The buccal membrane has seven ribs and marginal projections, as usual in the family Histiotectidae, and three dorsal ribs out of the seven are connected by double ligaments with the lateral sides of the bases of arms, the remaining four being attached by only a single ligament. The inner surface of the buccal membrane is finely wrinkled and of a purplish color.

The umbrella and protective membranes of the arms occur nearly in the same way as those of *M. hoylei* PFEFFER. The membranes of the umbrella between the two dorsal pairs of arms are fairly wide, extending about six or seven mm. up from the bases of the arms, while between the lateral arms, as well as between the third and fourth arms, they are very narrow, and between the ventral pair they are entirely wanting.

The protective membranes of three dorsal pairs of arms are of the usual breadth equally on both sides of the sucker-bearing surface, except at the proximal parts of the arms where they widen, making connective membranes between the bases of the arms, and imitating the umbrella as it occurs in *M. hoylei*, and the membranes of the specimen now before me extend about thirteen mm. up from the bases of all the arms, all the protective membranes of the ventral pair being very narrow.

The arm-suckers also resemble those of *M. hoylei* in shape and arrangement. They are small and globular, arranged clearly in two series. These two series, in the three dorsal pairs of arms, from their bases to a point about half-way to their tips, are widely separated, the distribution of the suckers in

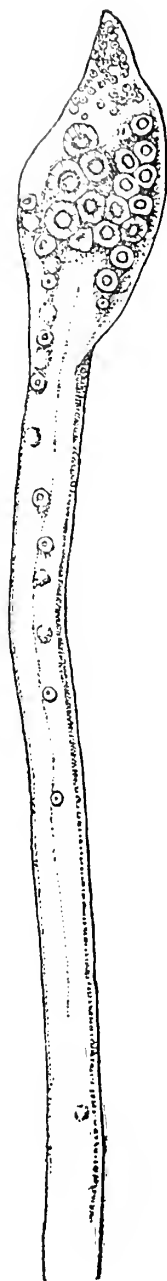


Fig. 2.—*Melcagroteuthis separata* sp. nov.  
distal half of left tentacle,  $\times 7$ .

each series on this part being very thin. But from the middle of the arms to their tips the two series are close to each other, the distribution becoming very thick. The suckers of the right ventral arm are a little smaller than those of the three dorsal pairs and they are arranged from the extreme base of the arm, in a somewhat thicker series than those of the other arms. The left ventral arm shows many different features from all the others, this being probably due to deformity.

The tentacles are slender, being much longer than twice the mantle-length. The stalks show nearly the same character as in *M. hoylei*, being provided with a flat inner surface, but the lateral boundary edges of this surface are not so distinct as to make "*Faden*" of PFEFFER'S (1912, p. 292) marked by grooves. The clubs at the distal end occupy about one-tenth the length of the tentacle, and are expanded into a spindle-shape. The fact that the two clubs show somewhat different features is probably due to the shrinkage of one of them (text-fig. 2).

The tentacular suckers show the principal characteristics of the present species, distinguishing it from *M. hoylei*. They are clearly divisible into three kinds by their situation: (1) Connective suckers, comprising from nine to ten suckers and from eight to ten fixing tubercles arranged in a single series, which begins on the ventral inner surface at a point about two-thirds from the base of the tentacle, and running diagonally into the dorsal side of the carpal portion, then going farther up to the middle part of the club along its dorsal margin. (2) Suckers of the hand portion, unequal in size, arranged in four longitudinal series; the most ventral series, comprising seven or eight suckers of

small size but a little larger than the connective suckers the next ventral series, composed of five suckers as small as those of the most ventral one ; and the third series, which is just median in the club, having three suckers a little larger than any of those of the preceding series. The most dorsal series, which is next to the connective sucker series in the hand portion, has only two or three suckers which are the largest amongst the tentacular suckers, being twice or thrice as large as those of the most ventral series. (3) Suckers of the distal portion are all minute, being much smaller than the connective suckers, and they are not connected to the sucker group of the hand portion as in *M. hoylei*, but are entirely separated from it<sup>1)</sup>, and they are distributed in a very irregular manner, except for their proximal ones which seem to be arranged more or less regularly in five or six series (text-fig. 2).

The tentacular suckers are of much depressed shape, their aperture being comparatively larger than those of the arm-suckers, and they have, along the margin of the aperture, distinct radial muscles, which are most distinctly visible on the largest suckers. The denticulation of the horny rings does not admit of being examined on account of the aperture being filled with the same horny substance as occurs in the arm-suckers.

The series of the horny tubercles runs along each outer median line of the three dorsal pairs of arms as well as along the median line of the dorsal surface of the mantle as occurs in *M. hoylei*. The tubercular series of the first pair of arms is the longest, and beginning at the anterior part of the dorsal surface of the head, it reaches a point a little proximal from the middle of the arms. It comprises ten or eleven large tubercles and some minute ones. The larger ones diminish in size towards the distal end of the series, the most proximal being the largest, and the smaller ones are intercalated among the larger. The series of the second pair is shorter than that of the first pair, beginning at the level of the anterior margin of the umbrella or a little posterior to it, and the number of tubercles in this series is ten or eleven. The series of the third pair is the shortest, with eight or nine tubercles.

1) Because of this characteristic, the specific name "*separata*" has been given to it.

The tubercular series of the dorsal surface of the mantle is not quite median in position, but deviates into the right side posteriorly. It begins at the projected middle point of the anterior margin of the mantle, and runs straight backwards for some distance; then it curves gradually into the right side, almost reaching the anterior end of the attachment of the right fin. The tubercles are not so many as thirty, as in *M. hoylei*, but only seventeen and are small, being nearly of equal size.

The luminous organs are uniform in appearance, thickly covering the ventral surface of the whole body and are found in less number on the dorsal surface.

On the ventral surface of the mantle, they are thickest in distribution at the anterior region, becoming thinner and diminishing in size towards the posterior end, and they show a regular arrangement in an oblique direction at the anterior half of the mantle. The most anterior transverse row along the edge of the mantle-opening consists of about thirty-eight organs, but of these only about twenty-five are visible from the ventral side. And when they are counted longitudinally along the median line of the ventral surface, though they do not show a quite regular series, the number is about twenty-four.

The dorsal surface of the mantle is provided with a small number of the organs, which show a most irregular arrangement and are asymmetrical in distribution on both sides, those of the left side being a little thinner in distribution and smaller in size than those of the right side. When the mantle is seen from the right side, the organs show a quite regular arrangement, the series diverging somewhat from the median part of the ventral anterior margin towards the posterior dorsal direction.

The ventral surface of the head is also thickly covered with organs arranged in oblique series. The most posterior transverse row along the anterior boundary edge of the neck consists of twenty-four organs, which is thickest in arrangement in the middle. The number of the organs in the row between both the eye-openings can not be counted, on account of the skin being torn, but judging from the arrangement of the anterior as well as the



posterior regions, it seems to be composed of about twenty organs, as occurs in *M. hoylei*.

The lateral regions of the dorsal surface of the head are provided with organs much smaller and much thinner in distribution than those of the ventral surface. The arrangement is irregular and asymmetrical on both sides, the organs of the right side being more numerous than those of the left side. When the head is seen laterally, the size and distribution of the organs differ greatly between the right and left sides: on the right side, the organs are numerous, becoming rarer towards the dorsal side, and there is found a longitudinal zone entirely free from organs, crossing the eye-opening and extending from the angle between the third and fourth arms to the neck. The margin of the eye-opening of this side is provided with twenty-four organs around the whole edge. The left side of the head has very few and minute organs, which are entirely lacking around the margin of the eye-opening.

The series of the luminous organs of the arms are only the continuation of those of the head, being arranged in some longitudinal series. The outer surface of the right ventral arm is thickly covered with the organs. There are nine series of the organs at the extreme base, which become reduced to six in the middle and to one at the extremity, the series which reaches to the extremity being the second one among the nine series from the ventral side.

On the proximal part of the third pair of arms, there are two series of the luminous organs which are all situated in a more ventral position than the median tubercular series, and the longest one of the two, comprising about forty organs, reaches the extreme end of the arm. The dorsal side of the median tubercular series has only a few organs of small size.

The ventral side of the tubercular series on the second pair of arms is provided also with two series of the luminous organs at the proximal part. The longer series of the two, comprising about thirty-three organs, reaches the extremities of the arms. The organs on the dorsal side of the tubercular series show nearly the same character in distribution as those on the equal surface of the third pair, but much less frequently. The organs of the first

pair are smaller and much rarer in occurrence than those of all the preceding arms, arranged very irregularly.

**Ommastrephes volatilis** sp. nov. (Pl. IV., figs. 1-6)

Local name: *Tobi-ika* (Sagami Prov.).

*Type*.—(I) One male and female specimens, taken by Mr. K. AOKI off Atami, Sagami Prov. on June 24, 1906.

(II) One male and two female specimens, caught by the same collector in the same part of the sea on Sept. 23, 1905. According to the information of the collector, they flew from the sea into the sky and colliding with the sail of his boat, fell down into it.

The following table shows the measurements of these five specimens (in alcohol).

|                          | No. I |       | No. II |       |      |       |       |
|--------------------------|-------|-------|--------|-------|------|-------|-------|
|                          | ♂     | ♀     | ♂      | ♀     | ♀    |       |       |
|                          | mm.   | mm.   | mm.    | mm.   | mm.  |       |       |
| Dorsal length of mantle  | 213   | 208   | 152    | 148   | 148  |       |       |
| Ventral length of mantle | 210   | 205   | 149    | 145   | 145  |       |       |
| Circumference of mantle  | 98    | 93    | 75     | 82    | 73   |       |       |
| Breadth of mantle        | 34    | 33    | 27     | 28    | 27   |       |       |
| Length of head           | 41    | 40    | 33     | 54    | 30   |       |       |
| Breadth of head          | 35    | 34    | 28     | —     | 26   |       |       |
| Length of fins           | 125   | 112   | 85     | 87    | 90   |       |       |
| Total breadth of finns   | 100   | 100   | 75     | 82    | 73   |       |       |
|                          | left  | right | left   | right | left | right | right |
| Length of first arm      | 110   | 108   | 73     | 76    | 73   | 73    | 72    |
| „ „ second arm           | 112   | 117   | —      | 99    | —    | 85    | 85    |
| „ „ third arm            | 115   | 114   | 109    | 108   | 86   | 85    | 85    |
| „ „ fourth arm           | 103   | 95    | 100    | 100   | 85   | 85    | 75    |
| Length of tentacle       | 187   | 194   | —      | —     | 180  | 180   | 143   |
| „ „ club                 | 60    | 63    | —      | —     | 55   | 55    | 50    |

The body is fleshy, with smooth skin. The mantle is very slender, the length being about six times the breadth, and it is conical, the broadest part

being the anterior, and has a very narrow posterior portion; the anterior margin is truncate, with neither projection nor emargination. The fins are long and are both together cordate with pointed lateral edges, the post-lateral edges being arched, and attenuated posteriorly, vanishing at some distance (13 mm. in a male with 213 mm. mantle-length) before the extreme end of the mantle (Pl. IV, fig. 1).

The head is as broad as the mantle-opening, with a sharp boundary edge behind. The eye-openings have, at a point a little below the middle of the anterior edge, a deep sinus, of which the dorsal margin is greatly thickened. The siphonal groove is fairly deep, being bounded by a distinct edge, the post-lateral portions of which, projecting a little, embrace the siphon laterally. The foveola in front is distinct, being bounded by a falcate fold posteriorly, but without any other folds either inside or outside. The olfactory crest of each side consists of one transverse and three longitudinal folds, the latter ones extending between the former and the posterior boundary edge of the head (Pl. IV, fig. 2).

The siphon, tapering a little forwards, extends generally to the middle of the head; the collar-like portion of the base is free posteriorly around the viscera, except at the nape where it joins the skin. The siphon is connected with its groove, by two pairs of ligaments, one pair of which is dorsal and the other entirely lateral in position. From between the dorsal pair there projects, in all the specimens examined, a muscular protuberance with a round head.

The arms are thick, long, and of subequal length, the formula being  $3 \div 2 > 1 > 4$ ; the longest arm is about half as long as the mantle. The first pair of arms is nearly quadrangular in section at the proximal halves and nearly cylindrical at the remaining distal halves. The second pair is also quadrangular either at the proximal parts or at the distal, and is provided, along the ventral outer edges, with a web as broad as its own protective membranes. The third pair is compressed laterally, being provided, along the whole length of the outer surfaces, with a very high keel, of which the highest

is in the middle, the height being much greater than the thickness of these arms. The fourth pair is quadrangular along the whole length, and has a broad web along the dorsal outer edge. The ventral protective membranes of the second and third pairs, are a little broader than the length of the suckers, but the dorsal membranes of these arms are about equal to their length. Both the protective membranes of the remaining arms are much narrower than all the preceding ones.

The arm-suckers are semicircular and arranged in two series. The size does not show any sexual dimorphism, but varies in the different arms of each individual, the suckers of the second and third pairs of arms being much larger than those of the first and fourth pairs: in a female of 208 mm. mantle-length, 3.2 mm. in I pair of arms, 3.5 mm. in II and III, 2.7 mm. in IV.

The horny rings of the arm-suckers have several distinct teeth along the distal margins; the proximal margins project slightly, forming a narrow border of smooth edge; the shape of the teeth varies in the different suckers: in the third pair of arms, the horny ring of one of the suckers in the second row from the base has ten somewhat pentagonal teeth thickly arranged, of which the lateral ones are much broader than the median and are very oblique; the horny ring in the sixth row has eight strong acute conical teeth, but of these the marginal ones are both obliquely quadrangular and much broader than the others; besides these, there are found five flat semicircular or quadrangular supplemental teeth, alternating thickly with the preceding ones, (Pl. IV, fig. 3a); the horny ring of the fifteenth row has eight slender and acute teeth, of which the marginal ones are a little broader than the remaining ones, and of which the interdental spaces are a little broader than those of the fore-going suckers (Pl. IV, fig. 3c); the horny rings of the remaining suckers, except those more distal than the fifteenth row, show a character intermediate between those above described. The denticulation of all the suckers more distal than the fifteenth row resembles closely that of the fifteenth row, but the number becomes less towards the extremities of the arms.

The hectocotylus is always in the right ventral arm, about two-thirds of the distal part showing the modification (Pl. IV, fig. 1a). Along two-thirds of the proximal part, there are thirty-four suckers arranged in two series; the middle three or four in each series being smaller than the remaining ones, especially with regard to those of the ventral series. The remaining distal part has no normal suckers but fifty sucker-bases, swollen into a transverse membranous shape, and arranged in two series, those of the ventral series being much smaller than those of the dorsal, and each sucker-base being provided with a minute papilla on its top. All these sucker-bases are connected with a median ridge running along between the two series. The distal two-thirds of the ventral protective membrane is enlarged and much thickened with a sculpture on the outer surface; the sculpture is composed of pits and grooves. There are found along the median line of the outer surface about fourteen pits, each sending sideways two grooves. Besides these, many small round or oval depressions and shallow grooves are found, being arranged in a somewhat regular manner (Pl. IV, fig. 4b).

The tentacles are about as long as the mantle, though they show some variation in length, owing probably to some external cause; the stem is a little compressed laterally and nearly quadrangular in section, the inner surface being flattened, with a web running from the base along the dorsal outer edge, extending to the outer surface of the club, where it becomes a little wider and bends towards the dorsal side distally. The clubs, occupying about one-third the whole length of each tentacle, are expanded lanceolately and show a triangle in section, with protective membranes about as broad as the length of the suckers. The suckers of the carpal portion are about twelve in number, arranged sparsely in two or three series, extending downwards to the exact middle of the tentacle, while all the remaining suckers are distinctly in four series. Ten central suckers of the hand portion are about thrice as large as the marginal ones in diameter; the suckers of the distal portion are smaller than the marginal ones of the hand portion and are arranged in about twenty-seven rows and in four series, of which the more

dorsal ones are always smaller than the more ventral. The suckers are nearly semicircular with a broad aperture, but the larger ones, having a very deep cavity, are longer in profile than the smaller suckers, which are shorter, with bottoms raised up to the level of the aperture. The horny ring in the largest sucker has, along the whole margin, about twenty sharp, slightly recurved conical teeth of about equal length, their breadth being a little narrower than the interdental spaces. Those of the marginal suckers show nearly the same characters in denticulation as the largest one, but the teeth of the lower margin are distinctly shorter than those of the upper. Those of the distal and carpal suckers also have teeth resembling those of all the preceding ones in the upper margin, but the edges of their lower margins show an even contour, projecting slightly and forming narrow borders.

The umbrella is very narrow. The buccal membrane has seven ribs, projections, and connectives, of which the dorsal-most is divided into two branches near the arm-bases, the inner surface being papillate. The outer lip is thin: the inner, thick and papillate. The so-called aquiferous pores between the arms and buccal membrane are four in number, between the first and second, and between the third and fourth, pairs.

The radula comprises seven series of somewhat thick teeth, the median tooth being tricuspid, the lateral bicuspid, and both the marginals unicuspid. The lateral teeth are about as long as the median, and the outer marginal a little longer than the combined length of the two succeeding medians (text-fig. 3).



Fig. 3.—*Ommastrephes volatilis* sp. nov.  
camera drawing of radula,  $\times 60$ .

The gladius is shorter than the mantle-length and is composed only of rhachis, lacking lateral vanes, but at the posterior part, narrow, wing-like laterai expansions from the rhachis are found, the length of which are about one-fifth of the whole length, making a shallow cone posteriorly (Pl. IV, fig. 6).

The present species resembles in many respects all the species of *Ommastrephes* hitherto known (*O. sagittatus*, *O. hawaiiensis*, *O. sloani*), to which it seems to stand in the nearest relationship among the Oegopsida, especially is this so with regard to the fact that the hectocotylus is only in the right ventral arm, and the siphonal groove has the foveola in front, bounded posteriorly by a crescentic pocket-like fold without any folds outside. Characters distinguishable from these species are shown in the following key, which was made by means of a comparison of the specimens whose mantle-lengths are about 148–215 mm.

1. Foveola of siphonal groove with several longitudinal folds within. Breadth of mantle about  $18-30\%$  of its length. Fins together distinctly broader than their length. Horny rings of arm-suckers provided with several sharp teeth on their distal margins, without any quadrangular supplemental teeth alternating with the preceding ones. Horny ring of largest tentacular sucker generally with quadrangular supplemental teeth alternating with sharp, ordinary teeth. Hectocotylus showed only by the degeneration of suckers and the swelling of the sucker bases .....

..... *O. sagittatus*, *O. hawaiiensis*, *O. sloani*.

2. Foveola of siphonal groove without longitudinal folds within, but smooth. Breadth of mantle  $16-19\%$  of its length. Fins distinctly longer than their total breadth, attenuated posteriorly. Horny rings of arm-suckers varying in denticulation in different suckers of each arm; largest sucker of II arm provided with quadrangular supplemental teeth alternating with long, sharp, ordinary teeth. Horny ring of largest tentacular sucker with only sharp, and without any supplemental, teeth. Hectocotylus shown not only by the degeneration of suckers and the swelling of sucker bases, but also by the thickening and enlarging of ventral protective membrane and by sculpture composed of pits and transverse grooves on outer surface .....

..... *Ommastrephes volatilis* n. sp.

**Symplectoteuthis luminosa** sp. nov. (Pl. IV., figs. 7-13).Local name : *Suji-ika* (Sagami Prov.)

*Type*.—Three male and seven female specimens, taken six miles off Misaki coast, in 700 fathoms, Aug. 5, 1906. Their measurements are as follows :

|                          | ♂<br>mm. | ♂<br>mm. | ♂<br>mm. | ♀<br>mm. | ♀<br>mm. | ♀<br>mm. | ♀<br>mm. | ♀<br>mm. | ♀<br>mm. | ♀<br>mm. |
|--------------------------|----------|----------|----------|----------|----------|----------|----------|----------|----------|----------|
| Dorsal length of mantle  | 143      | 154      | 166      | 120      | 122      | 141      | 151      | 151      | 155      | 157      |
| Ventral length of mantle | 138      | 151      | 191      | 116      | 120      | 139      | 147      | 148      | 151      | 155      |
| Circumference of mantle  | 60       | 92       | 102      | 75       | 82       | 86       | 97       | 98       | 92       | 97       |
| Breadth of mantle        | 31       | 33       | 35       | 24       | 28       | 28       | 32       | 33       | 33       | 33       |
| Dorsal length of head    | 27       | 33       | 34       | 19       | 31       | 28       | 33       | 32       | 32       | 32       |
| Breadth of head          | 30       | 34       | 34       | 23       | 28       | 28       | 32       | 31       | 32       | 30       |
| Length of fins           | 69       | 72       | 83       | 52       | 54       | 66       | 71       | 76       | 75       | 78       |
| Total breadth of fins    | 76       | 80       | 90       | 62       | 72       | 73       | 90       | 87       | 81       | 85       |
|                          | L. R.    | L. R.    | L. R.    | L. R.    | L. R.    | L. R.    |          | L. R.    | L. R.    | L. R.    |
| Length of first arm      | 46 46    | 51 51    | 54 55    | 32 31    | — 45     | 43 43    | —        | 50 50    | 50 50    | 50 50    |
| „ „ second arm           | 53 53    | 57 55    | 60 59    | 36 36    | 49 49    | 50 50    | —        | 54 54    | 57 57    | 57 57    |
| „ „ third arm            | 50 50    | 55 55    | 56 56    | 35 36    | 47 47    | 49 49    | —        | 54 53    | 55 57    | 56 56    |
| „ „ fourth arm           | 48 50    | 53 55    | 56 60    | 36 36    | 47 47    | 49 49    | —        | 54 54    | 55 55    | 57 57    |
| Length of tentacle       | 68 70    | 75 75    | 96 96    | 60 60    | 68 68    | 70 70    | —        | 85 80    | — 63     | 100 102  |
| „ „ club                 | 32 32    | 35 35    | 40 38    | 23 23    | 32 32    | 31 31    | —        | 35 35    | — 32     | 38 38    |

The body is fleshy and the skin smooth. The mantle is conical, the broadest portion being in the anterior margin, and one-third of the posterior portion is more attenuated than usual. The emargination of the ventral anterior edge and the projection of the dorsal edge are both very slight. The fins which do not reach the extreme end of the mantle, are both together rhomboidal, with slightly concave latero-posterior edges, and are notched at the anterior attachment (Pl. IV, fig. 7).

The head is as broad as the mantle-opening, with a sharp boundary edge posteriorly. The siphonal excavation is deep and acuminate in front, being marked by a distinct boundary edge, of which the post-lateral parts project forming thick folds which embrace the siphon laterally. At the apex, it has a small rhomboidal foveola which has about eight faint longitudinal folds within, and outside the foveola, two or three small oblique folds are



seen in each side. The olfactory crest of each side consists of one transverse and three longitudinal folds, the latter stretching between the transverse fold and the posterior boundary edge of the head. Each eye-opening, at the middle of the anterior edge, has a distinct sinus, of which the dorsal margin is thickened (Pl. IV, fig. 8). The siphon tapers a little forwards, and reaches the middle of the head; the collar-like portion of the base is free posteriorly around the viscera except at the nape where it is fixed to the skin, and the ventral margin is highly arched with a thin membranous border. The siphonal resisting cartilages are triangular, with a spindle-shaped elevation in front; the groove is  $\perp$ -shaped, the longitudinal part curving towards the ventral side anteriorly. The middle portion of the cartilage is joined by the ligament to the mantle cartilage, and the posterior margin is also connected at the middle with the same. The dorsal connective ligaments of the siphon are in two pairs, one pair of which is median in position and the other lateral. The resisting cartilage of the nape is elongate and expanded anteriorly, with a pair of thick submedian ridges, which extend some distance backwards beyond the collar-like portion of the siphon, with another faint longitudinal ridge running between them.

The arms are subequal, the formula of length being about  $2\frac{1}{2} : 4\frac{1}{2} : 3 > 1$ ; the longest arm is about one-third the length of the mantle.

The first and second pairs of arms are about quadrangular in section; the former pair has a narrow web along each proximal half of both outer edges, while the latter pair is provided only along the ventral outer edge with a web, broader than the preceding ones and extending from the base of the arm up to the extreme end. The protective membranes of both sides of the first pair, as well as the dorsal membrane of the second pair, are a little narrower than the length of the suckers; while the ventral membrane of the second pair is slightly broader than the preceding ones, being as broad as the length of the suckers.

The third pair is compressed laterally, being provided along the whole median line of the outer surface, with a keel as high as the thickness of the

arm and highest towards a point one-third from the base of the arm. The ventral protective membranes are very broad, being about thrice as wide as the length of the suckers, while the dorsal membranes are narrower than the length of the suckers.

The fourth pair is nearly quadrangular, with a narrow web along the ventral outer edge, and a broad one along the dorsal, the proximal part of the dorsal web being as broad as the thickness of the arm; the protective membranes of both sides are alike about half as broad as the length of the suckers.

The arm-suckers are semicircular, with a broad aperture and oblique stalk, and they are arranged in two series; the horny rings have sometimes blunt, sometimes sharp, teeth on the distal margin, surrounded by a papillary area with numerous striations radially arranged. The horny ring of one of the suckers in the fourth row from the base on the second arm of a male whose mantle-length is 116 mm, has a large, thick, triangular tooth with acute point at the most distal edge, and on each side of it, seven flattened teeth somewhat thickly arranged and of much smaller size, some of these being blunt and some sharp; the horny ring of a sucker from the sixth row shows characters nearly equal to the preceding ring, but all the teeth of this ring have much blunter cusps, and the marginal tooth on each side is very much thicker, being as large as the median one (Pl. IV, fig. 9a); the horny ring of a sucker from the tenth row has, on the distal margin, nine teeth which are much narrower and sharper than those of the preceding rows (Pl. IV, fig. 9b).

The size of the suckers varies a little in the different arms of each individual, as the following table shows:

|                                              | ♂   | ♂   | ♀   | ♀   | ♀   |
|----------------------------------------------|-----|-----|-----|-----|-----|
|                                              | mm. | mm. | mm. | mm. | mm. |
| Mantle-length .....                          | 154 | 166 | 120 | 122 | 151 |
| Diameter of largest sucker of first arm..... | 2.1 | 2.5 | 1.3 | 2   | 2.5 |
| „ „ „ „ „ second arm .....                   | 3.4 | 3.8 | 2   | 2.5 | 3.5 |
| „ „ „ „ „ third arm .....                    | 2.5 | 2.8 | 1.7 | 2.2 | 2.8 |
| „ „ „ „ „ fourth arm .....                   | 1.8 | 2   | 1.2 | 1.5 | 2   |

The hectocotylus is in the left ventral arm, of which one-third of the distal portion shows modification to a greater degree ; the suckers of the proximal eleven rows are of normal size and appearance, and those of the remaining distal sixteen rows are very minute, without the usual shape of suckers, but papilla-like, with swollen bases (Pl. IV, fig. 10).

The tentacles show some individual variation in length, according to the state of preservation, but they seem to be about half as long as the mantle ; the stalk is a little compressed laterally and is nearly quadrangular in section, the inner surface being flattened and bounded laterally by sharp edges, which are continuous with the protective membranes of the club. The clubs are lanceolate, occupying from one-half to one-third the whole length of the tentacles, each having a web on the outer surface ; the continuation of the web begins as a ridge at each extreme base of the tentacles, and is expanded on the clubs, bending towards the dorsal sides distally. The suckers are arranged in four series and are unequal in size, eight or ten central suckers of the hand portion being much larger than the others. They are semicircular, with large apertures ; the papillary area has numerous horny striations of the same character as in the arm-suckers. In the carpal portion, two mammillary protuberances are found in the dorsal side of the sucker-bearing surface, and the suckers which exist more proximally than those protuberances, show an individual variation in number from two to eight. The largest central sucker of the hand portion is about as large as the marginal ones twice in diameter, and is about equal to the largest sucker in the arms ; this sucker has, in all the specimens examined by me, only a single triangular tooth of large size on the distal-most edge of the horny ring (Pl. IV, fig. 11a) ; the marginal suckers have about twenty triangular teeth along the whole edge of the ring, sometimes with a number of supplemental teeth alternating with them (Pl. IV, fig. 11b).

The buccal membrane is finely wrinkled and has seven ribs, projections, and connectives ; the dorsal connective is divided into two branches near the bases of the first pair of arms, and has no membranous septum below, so

that all the aquiferous pores between the first pair of arms, as well as between the first and second arms of both sides, are continuous with each other. Besides the pores of the dorsal side, there are also found pores between the lateral arms, as well as between the third and fourth arms, but in some specimens, those between the lateral arms of the left side are not found.

The radula

comprises seven series of teeth; the median tooth being tricuspid, the lateral bicuspid and both marginals unicuspid; the outer marginal teeth are very narrow, the length

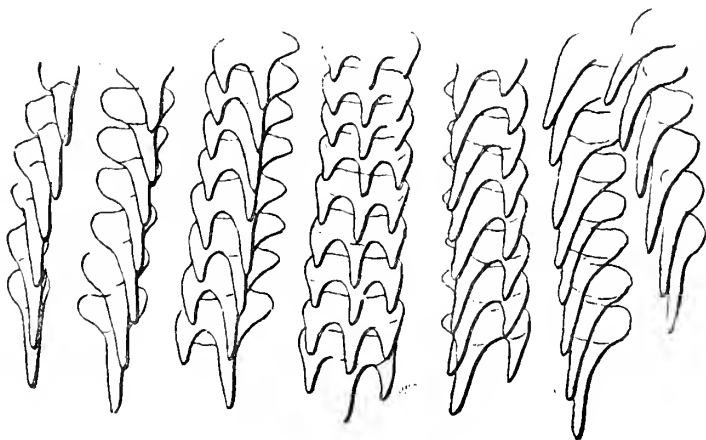


Fig. 4.—*Symplectoteuthis luminosa* sp. nov. camera drawing of radula,  $\times 60$ .

being equal to the combined length of the two succeeding median teeth (text-fig. 4).

The gladius is very narrow, without lateral vanes, but at the posterior end, there is a slightly expanded portion, the length of which is about one-fifth of the whole, and which forms a cone posteriorly (Pl. IV, fig. 13).

On the ventral surface of the head, a paler macula is found at the base of each ventral arm, and on the ventral surface of the mantle, there is also a pair of longitudinal zones of the same character, which runs along the whole length of the mantle, dividing laterally the same surface into three equal areas, and, curving slightly outwards anteriorly, enlarged at the extreme anterior ends. Those maculae and zones seem, judging from their histological structures, to be phosphorescent organs. When the mantle is flayed, a long white substance which has always another brownish substance along the median line, appears along each paler zone. The white substance is not con-

tinuous along the whole length, but is divided into three pieces: the foremost of oval shape, is the smallest, the middle is the longest, stretching from a little behind the anterior mantle-margin to about the level of the middle of the fin, and the last is about eight times as long as the first, and does not reach the extreme end of the mantle but is one length distant from it.

Under the skin of the paler maculae on the head, is also found the same substance as seen in the paler zones on the mantle, but it is a little thicker and is transversely oblong. Such substances of the mantle, as well as of the head, are all situated in the depressions of the muscles at those places, and the skin covering them is a little thicker than that covering the remaining parts (Pl. IV, fig. 12).

The principal differences of the present species from *S. oualaniensis* (LESSON), with which it stands in the nearest relationship, are as follows:

1. Luminous organ absent. Hectocotyliised left ventral arm much thicker and longer than right ventral arm, with 14 suckers and broad protective membranes greatly thickened; about  $\frac{3}{5}$  of the distal part of whole length is bare but only with a narrow longitudinal ridge. Horny ring of largest tentacular suckers with a large tooth in each of 4 corners and 5 small teeth between each larger succeeding 2.....*Symplectotenthis oualaniensis* (LESSON).

2. Luminous organ present. Hectocotyliised left ventral arm a little shorter than right ventral one, and as thick as the latter, and provided with about 24 normal suckers along proximal part; protective membranes as broad and thick as those of right arm; distal  $\frac{1}{3}$  of whole length not bare but with 34 minute tubercles each on a swollen base. Horny rings of largest tentacular suckers with only a large tooth on upper-most edge .....  
..... *Symplectotenthis luminosa* sp. nov.

**Explanation of Plate IV.**

*Ommastrephes volatilis* sp. nov. —

Fig. 1. Dorsal view of male,  $\times 1/2$ .

Fig. 2. Lateral aspect of the same,  $\times 1/2$ .

Fig. 3. Camera drawing of horny rings from third left arm,  $\times 30$ . a) From sucker of sixth row from base of arm; b) from sucker of tenth row; c) from sucker of fifteenth row.

Fig. 4. Hecotocotylus,  $\times 1/2$ . a) Inner aspect; b) lateral aspect showing sculpture.

Fig. 5. Camera drawings of oblique-lateral view of horny ring from largest tentacular sucker,  $\times 13$ .

Fig. 6. Ventral aspect of gladius,  $\times 1/2$ .

*Symplectotenthis luminosa* sp. nov. —

Fig. 7. Ventral view of male,  $\times 1/2$ .

Fig. 8. Lateral aspect of head of female,  $\times 1/2$ .

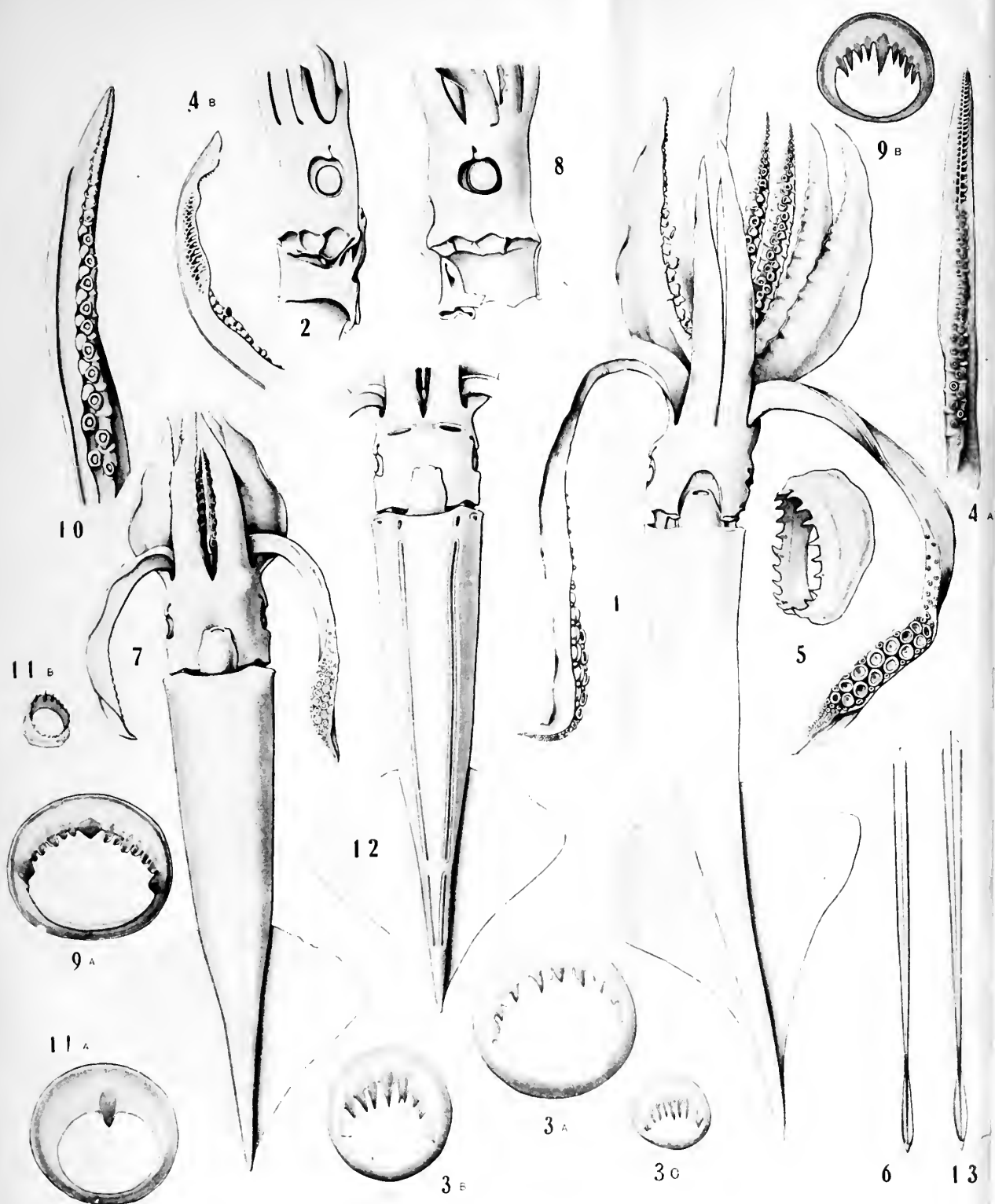
Fig. 9. Camera drawings of horny rings from second left arm,  $\times 30$ . a) From sucker of sixth row from base of arm; b) from sucker of tenth row.

Fig. 10. Inner aspect of hectocotylus,  $\times 5/4$ .

Fig. 11. Camera drawings of horny rings from tentacle,  $\times 30$ . a) From largest sucker in inner row; b) from marginal sucker in hand portion.

Fig. 12. Ventral aspect of body of male showing luminous organs after flaying,  $\times 1/2$ .

Fig. 13. Ventral aspect of gladius,  $\times 1/2$ .



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APR 29 1924

# BEITRÄGE ZUR KENNTNIS DES KEHLSACKES BEIM RENNTIERE

von

**Kotaro Ogura und Jinshin Yamane**

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Der sog. Kehlsack, ein zwischen Schildknorpel und Zungenbein sich ausdehnender Nebenraum des Larynx, wurde beim Renntiere zuerst von P. CAMPER gefunden.<sup>1)</sup> Weitere Einzelheiten darüber wurden jedoch erst von E. LÖNNBERG eingehend geschildert.<sup>2)</sup> Dieser Autor hat durch seine Untersuchungen, die er an mehreren aus Lappland erhaltenen Kehlköpfen durchführte, über das Vorkommen und die Ausdehnung dieses Organs etwelche Klarheit geschaffen. Da aber die genaueren anatomischen Verhältnisse bzw. die physiologische Bedeutung des Kehlsackes bis heute noch unklar geblieben sind, so sind wir an weitere Untersuchungen herangetreten.

Unser Material war allerdings nicht sehr umfangreich. Zur makroskopischen Anatomie diente uns ein bei der postmortalen Untersuchung gewonnener und in Alkohol konservierter Kehlkopf, der von einem männlichen Renntiere stammte, das im Jahre 1912 von Japanisch-Sachalin an unsere Universität geschickt worden war, aber bald nachherdaselbst einging. Zur mikroskopischen Untersuchung lag uns ein von einer Renn-tierkuh stammender Kehlsack vor, der von dem einen der Autoren auf seiner Exkursion durch Japanisch-Sachalin im Jahre 1914 von einem Oroken erworben und in mehrmals gewechselter Müller'scher Flüssigkeit zurückgebracht worden war.

Wenn man die Kehle des Renns ventral öffnet, so findet man zunächst

---

1) Nach LÖNNBERG, s. u.

2) LÖNNBERG, E., Anat. Anz., Bd. XXI, (1902) pp. 467-474.

einen ovalen Sack von ca 11 cm Länge und 8 cm Breite, der locker am Unterhautsbindegewebe anliegt und sich mit Ausnahme des unteren Teiles des Ringknorpels über den ganzen Kehlkopf ausdehnt. Darunter liegt beiderseits der *Musculus sternohyoideus*. Dieser ist seitlich am Kehlsack befestigt, aber durch das Bindegewebe getrennt, sodass kein inniger Zusammenhang zwischen beiden zu finden ist. Ein von CAMPER abgebildeter Muskel, den wir bei LÖNNBERG erwähnt finden, muss wohl dieser sein.<sup>1)</sup> Nach Beseitigung des *M. sternohyoideus* bemerkt man die zwei bandartigen Muskel, *M. sternothyreoideus* und *M. hyothyreoideus*. Unter dem *M. hyothyreoideus* liegt der sehr breite *M. thyreopharyngeus*. Alle diese Muskeln stehen in keiner Beziehung zum Kehlsack.

Der Kehlsack ist dorsal auf einer gewissen medianen Strecke sehr dicht mit dem Perichondrium des Schildknorpels verwachsen, während die Verbindung mit der *Membrana threohyoidea* locker ist. Der Kehlkopf unseres Präparats misst dorsal vom vorderen Bogen des Giesskannenknorpels zum dorso-caudalen Rand des Ringknorpels 105 mm, und ventral von der *Incisura thyreoidea oralis* zum ventro-caudalen Rand des Ringknorpels 95 mm. Der grösste dorso-ventrale Durchmesser beträgt 60 mm. Die Epiglottis ist 50 mm breit und 30 mm lang. Vorne ist sie ca 5 mm tief eingekerbt. Die Schildknorpellammellen sind gänzlich verknöchert. Die Stimmbänder sind ca 40 mm lang. Aboral von diesen findet sich eine ca 35 mm lange Vertiefung. Etwa 12 mm vorwärts von dieser Vertiefung und zwar an der Basis des Kehlsackes liegt ein fast ovales Loch von ca 12 mm Länge und 8 mm Breite. Das Loch führt am Vereinigungswinkel der beiden Seitenplatten des Schildknorpels und zwar an der Basis der Epiglottis in den Kehlsack hinein.

Der Kehlsack ist mit Schleimhaut ausgekleidet, deren Oberfläche mehr oder weniger mit seröser Flüssigkeit befeuchtet ist. Die Schleimhaut der seitlichen und ventralen Wände ist sehr faltenreich, diejenige der dorsalen dagegen nahezu eben und glatt. Stellenweise findet man schon zahlreiche, mit blossen Auge bemerkbare Drüsenpakete von ca 5 mm Länge und 2 mm

---

1) LÖNNBERG, l. c.

Breite. Am häufigsten sind diese aber in der Nähe von dem eben beschriebenen Loch. Die Wand des ganzen Sacks ist auch mit zahlreichen Gefässen versehen.

Das Epithel der Schleimhaut ist anfangs niedrig, dann höher geschichtet und schliesslich Zylinderepithel. Das aus zahlreichen elastischen Fasern und aus fibrillärem Bindegewebe bestehende Stratum proprium ist reichlich mit Leukozyten eingelagert; es enthält auch vereinzelte Lymphknötchen.

In der Submucosa befinden sich die zusammengesetzten, alveotubulösen Drüsen, die von der umgebenden Bindegewebshülle in verschieden grosse Komplexe geteilt sind. Sie bilden zuweilen, wie schon erwähnt, grosse Pakete. Besonders drüsenreich sind die ventralen und seitlichen Wände, die wirbelseitige Wand dagegen ist auf eine gewisse mediane Strecke weit drüsenlos. Die straffe Submucosa ist die Trägerin grösserer Gefässstämme. Sehr bemerkenswert ist, dass die quergestreiften Muskelfasern stellenweise in der Submucosa vorkommen. (Tafel V). In der dorsalen Wand fehlen sie ganz; sie treten, grössere oder kleinere Muskelbündel bildend, mehr oder weniger in der seitlichen Wand auf. Vorwiegend kommen sie aber in der ventralen Wand vor.

Die äusserste Schicht des Kehlsacks ist eine Fettgewebsschicht, zuweilen eine Bindegewebsschicht von lockerem Bau.

Was nun die Funktion des Kehlsackes anbetrifft, so hat sich schon C. GEGENBAUR dahin ausgesprochen, dass die durch verschiedene Modifikationen der Struktur des Kehlkopfs bedingten Nebenhöhlen gewisser Tiere als Resonanzapparate zur Verstärkung der Stimme dienen.<sup>1)</sup> Derselben Ansicht ist LÖNNBERG.<sup>2)</sup> Er meint, der aufgeblasene Kehlsack werde die verknöcherte Thyreoidealammelle oben und unten von luftgefüllten Höhlen umgeben und in dieser Weise zur Resonanz beitragen. LÖNNBERG nimmt ferner an, dass die Schleimhautfalten, die das in den Kehlsack führende Loch umgeben, möglicherweise auch derart gespannt werden, dass sie bei der Schaller-

1) GEGENBAUR, C., Vergleichende Anatomie der Wirbeltiere, Bd. II. (1901) p. 297.

2) l. c.

zeugung von Bedeutung sein können. Vom anatomischen Standpunkte aus unterliegt es keinem Zweifel, dass der Kehlsack als Resonanzboden für die erzeugten Töne dient. Ebenso ist gewiss, dass dieses Organ selbst zur Tongebung gewissermassen beiträgt. Von grösster Bedeutung für den Mechanismus der Tongebung sind die von uns gefundenen Muskelfasern. Die Wirkung dieser Muskelfasern, deren Bewegungen wahrscheinlich bis zu einem gewissen Grade der Willkür unterworfen sind, besteht darin, dass sie die schwingende Bewegung der membranartigen Kehlsackwände bei jedem Aufblasen des Sackes begünstigen, wodurch die dem Renntiere eigentümlichen groben grunzenden Töne verursacht werden.

Es fragt sich nun aber auch, auf welche Weise der Kehlsack aufgeblasen wird. Unserer Beobachtung nach muss dies durch die aus den Lungen getriebene Luft geschehen, denn das in den Kehlsack führende Loch liegt fast vertikal, weshalb es unmöglich ist, dass die von dem Maul und den Nasenlöchern aufgenommene Luft direkt hindurch passiert. Wie unser anatomischer Befund zeigt, spielen die Halsmuskeln bei der Funktion des Kehlsackes keine Rolle.



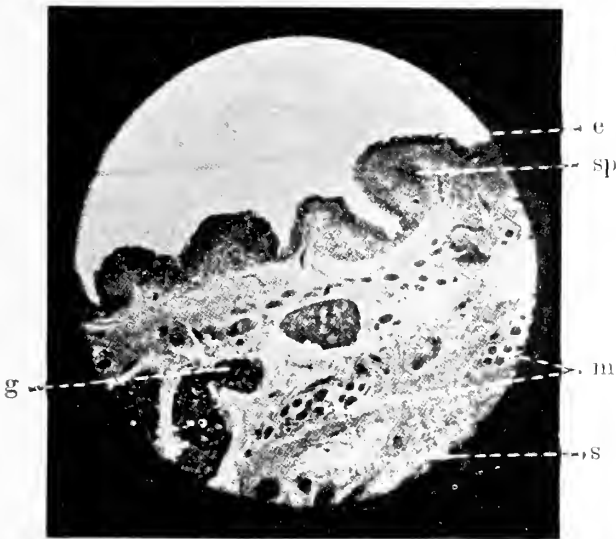


Fig. 1

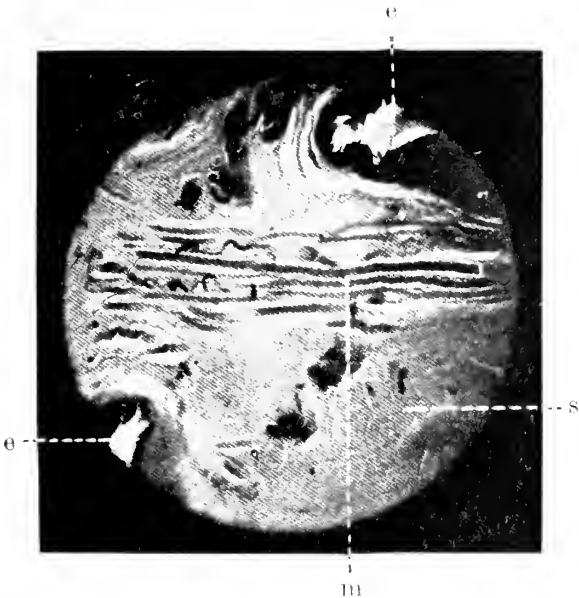


Fig. 2



**Erklärung der Tafel V.**

Fig. 1. Querschnitt durch die Schleimhaut an der ventralen Wand des Kehlsackes einer Rennkuh. Vergr. ca 30 : 1.

*e* Schleimepithel; *sp* das mit zahlreichen Leukozyten eingelagerte Stratum proprium; *g* die zusammengesetzten alveo-tubulösen Drüsen; *m* Muskelbündel; *s* Submucosa.

Fig. 2. Flachschnitt durch dieselbe. Vergr. ca 30 : 1.

# ÜBER EINE SOGENANNT E ENDOPARASITÄRE ACARINA

von

**Kotaro Ogura** und **Koichi Ichikawa**



Bis jetzt ist das Vorkommen einer Acarina als wirklichem Endoparasiten nur ein einziges Mal und zwar im Jahre 1901 von J. de HAAN und G. GRIJNS<sup>1)</sup> mitgeteilt worden, indem diese beiden Forscher bei der Sektion eines Cynocephalus (von Süd-Sumatra) im Lungenparenchym den eingangs erwähnten Parasiten entdeckten. Doch ist die Beschreibung nicht ausführlich genug, um diesen Parasiten mit Sicherheit als wirklichen Endoparasiten zu bezeichnen, besonders da alle parasitischen Acarinen des Menschen und der Haustiere als Ektoparasiten betrachtet werden. Einige Arten vermögen nun oft in den äusseren und mittleren Gehörgang oder in die Nasen- oder Mundhöhle einzudringen und dadurch, dass sie zufälligerweise in die Lunge gelangen und dort weiterleben, eine chronische Entzündung hervorzurufen. Damit sind wir aber noch nicht berechtigt, diesen Parasiten als Endoparasiten anzusprechen.

Die vorliegende Arbeit will deshalb zur Lösung dieser Frage einen Beitrag leisten. Als Untersuchungsmaterial diente den Verfassern ein in Japan geborener Macacus, in dessen Lungenparenchym sich eine beträchtliche Anzahl von Acarinen vorfand.

## **Allgemeiner Sektionsbefund des Falles.**

Ein in Japan geborener männlicher Macacus, der am 18. Dezember 1912 fast 24 Stunden nach dem Tode seziert wurde. Leider war die Kranken-

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1) HAAN, J. de u. GRIJNS, G., Eine neue endoparasitäre Acaride. (Centralbl. f. Bakt. Abt. I. Orig. Bd. 30. 1901)

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[Jour. of the College of Agr., Tohoku Imp. Univ., Sapporo, Vol. VI, Pt. 1, April 1915]

geschichte unklar. Die Sektion ergab folgenden Befund:

Eine stark abgemagerte männliche Leiche mit schwacher Muskulatur und stark atrophischer *Paniculus adiposa subkutaneus*. Die Haut trocken und blass, mit zahllosen Läusen am ganzen Körper. An der rechten Seite des Körpers schwache blassrötliche Totenflecken. Die Totenstarre schon ausgelöst.

a) **Bauchorgane:** In der Bauchhöhle keine abnorme Flüssigkeit enthalten. Das Grossnetz fettarm und trocken. Das Peritoneum trocken und matt. Unter den Bauchorganen keine abnorme Verwachsung und Verlagerung. Die Leber etwas verkleinert und blutarm, mit der ihr eigenen Organfarbe, sonst keine nenneswerte Veränderung. In der Gallenblase reichliche dicke gelbbraunliche Galle. Die Milz auch stark atrophisch mit stark vermindertem Volumen. Beide Nieren fast gleich beschaffen. Die Kapsel leicht abziehbar. Die Oberfläche glatt, mehrere kleine nadelspitzgrosse rötliche Pünktchen zeigend. Die Schnittfläche ziemlich blutreich und im allgemeinen matt. An der Rinde bemerkt man die obengenannten Pünktchen und am Mark mehrere rötliche Streifen. Das Becken etwas erweitert und dessen Schleimhaut ödematös. Der Magen gut retrahiert, eine kleine Menge von flüssigen Speiseresten aufweisend. Seine Schleimhaut ödematös mit dickem Schleim bedeckt. Die Pankreas blass und verkleinert.

Die Gedärme im allgemeinen ödematös und leicht injiziert, besonders auffallend am Dickdarm. Ihre Schleimhaut stark ödematös und mit dickem etwas gelblichem Schleim bedeckt.

b) **Brustorgane.** Die Pleura etwas matt, in den Höhlen ca 50 ccm einer gelblichen klaren Flüssigkeit enthaltend. Das Herz etwas klein; die Spitze ziemlich konisch, doch durch die linke Kammer gebildet. Das subepicardiale Fettgewebe sehr spärlich, und die Kranzarterie nicht geschlängelt. Myocardium bräunlich; die Klappen zart. Die rechte Herzhälfte und besonders die rechte Kammer ziemlich erweitert. Die Aorta thoracalis et abdominalis weist fast keine bemerkenswerten Veränderungen auf. Bei beiden Lungen die Pleura leicht verdickt und die Konsistenz etwas vermehrt. Dicht unter

der Pleura liegen mehrere reiskorn- bis submiliargrosse, grauweissliche und scharf von der Umgebung demarkierte Knötchen, die sich etwas auf der Pleura erheben. Beim Einschneiden finden sich mehrere der oben genannten Knötchen auch tief in dem Parenchym. Einige davon kommunizieren durch sehr feine Röhrenchen miteinander oder mit Bronchien. Die stark fibrös verdickten Wände der Knötchen und der Bronchien zeigen kleine Blutungsherde und sind samt ihrer Umgebung stark hyperämisch und fibrös induriert.

In den Lumen der Knötchen oder Bronchien befinden sich in geringer Anzahl nadelspitzgrosse weissliche körnige Massen, die an der Wand festhalten und sich nach schwacher Vergrösserung als eine Art Acarina erweisen. Doch das Vorkommen dieses Parasiten, sowie auch die obengenannten Veränderungen beschränken sich auf die feinen oder mittleren Bronchien, wogegen der Bronchus und dessen Hauptäste fast keine nennenswerte Veränderung zeigen, ausser der Schleimhaut, die etwas hyperämisch ist. Hilusdrüsen fast normal.

c) **Halsorgane.** Die Schleimhaut der Nasen- und Mundhöhle, des Larynx und der Trachea anämisch, sonst ohne Besonderheiten und keine Milben tragend. Sonstige Organe im allgemeinen atrophisch.

d) **Beckenorgane.** Die Rektalschleimhaut ödematös durchtränkt und mit dickem Schleim bedeckt. Die Harnblase gefüllt mit reichlichem gelblichem Harn von getrübler Beschaffenheit. Die Prostata und die Hoden zeigen fast keine bemerkenswerten Veränderungen.

### **Sektionsdiagnose:**

- 1) Beidseitige chronische katarrhalische Bronchitis und Peribronchitis.
- 2) Zahllose Läuse an der ganzen Körperoberfläche.
- 3) Beidseitige exudative Pleuritis.
- 4) Braune Atrophie des Herzens.
- 5) Leichte Dilatation der rechten Herzkammer.
- 6) Beidseitige parenchymatöse Nephritis.
- 7) Chronischer Magendarmkatarrh.
- 8) Atrophie der Pankreas, Milz und Leber.
- 9) Hochgradige Abmagerung und einfache Anämie.

**Mikroskopischer Befund mit besonderer Berücksichtigung der Frage, ob der vorliegende Parasit eine neue endoparasitische Acarina sei oder nicht.**

Das Material, das sowohl verschiedene Teile der äusseren Haut, als auch die Nasen-, Mund- und Trachealschleimhaut umfasst, wurde sorgfältig mit der Nadelspitze ausgekratzt. Zwecks Beantwortung der Frage, ob die Acarina vom eigenen Körper oder einem anderen Individuum aus durch Einatmen in die Lunge gelangt sein könne oder nicht, wurden etliche mit Jodkali-lösung durchsichtig gemachte Präparate mikroskopisch untersucht, doch mit negativem Erfolg, obschon derartige Acarinen schon bei sehr schwacher Vergrösserung, ja sogar bei scharfem Betrachten von blossen Auge sichtbar sind.

Da die Veränderungen, die durch die Acarina hervorgerufen werden, auf die Lunge beschränkt sind, so wurde diese in 4% iger Formollösung fixiert und in Paraffin eingebettet. Fast ein halbes Lättchen wurde ganz in Serien geschnitten. Die betreffenden Schnitte wurden auf verschiedene Weise gefärbt. Die Untersuchung ergab folgende Resultate

Die Pleura zeigt sich, besonders da, wo die obengenannten Knötchen auftreten, ziemlich stark fibrös verdickt, doch besteht keine Zellinfiltration. Diese Knötchen, sowie die erweiterten und verdickten Bronchiolen, in denen der Parasit schmarotzt, liegen dicht unter der Pleura oder auch tief im Parenchym, stehen miteinander oder mit den Bronchien und Bronchiolen oder auch mit den Alveolen in Verbindung, doch ist die Kommunikation häufig fast gänzlich aufgehoben. Die Grösse der Knötchen ist ganz verschieden. In der Wandung jedes Knötchens sind Glattmuskelbündel und Glattmuskelschichten nachweisbar. Die Höhle ist teilweise mit zerstörtem kubischem oder zylindrischem Flimmerepithel ausgekleidet. Ferner finden sich die Äste der Arteria pulmonalis dicht an der Wand. Gestützt darauf ist als sicher anzunehmen, dass die Knötchen nichts anderes als ektatische Bronchien oder Bronchiolen sind. Die mit Glattmuskelbündeln bekleideten Knötchen müssen die feinen Bronchien oder Bronchiolen und die mit Glattmuskelschichten bekleideten

Bronchien sein. Die Lumen der Bronchien und Knötchen, in denen der Parasit schmarotzt, sind stark erweitert und verdickt; oft erscheint die Schleimhaut hier und da zerstört, an einigen Stellen sind kleine Blutungsherde vorhanden; dicht unter der Schleimhaut liegt ganz wenig Bindegewebsschicht, ferner sind stark verdickte Glattmuskelbündel (an Bronchiolen oder feinen Bronchien) oder Glattmuskelschichten (an Bronchien) vorhanden; unter der Glattmuskelschicht bemerkt man granulierendes Zellinfiltrat, das schon bindegewebig induriert ist und zunächst faserigen Knorpel vortäuscht. Im Granulationsgewebe und dessen Umgebung befinden sich erweiterte und gefüllte Kapillaren, Venen und Arterien (zum Teil ziemlich hochgradig sklerotisch, obgleich die Aorta keine merkliche Sklerose zeigt); in dem Granulationsgewebe und dessen Umgebung befinden sich mehrere pigmenthaltende Zellanhäufungen; doch sind die Kerne nicht gut färbbar, das Plasma ist meist blasig in den Alveolen oder deren Reste verteilt; das Pigment reagiert nicht bei der Berlinerblaureaktion. Diese Veränderungen, die durch die blosse makroskopische Untersuchung festgestellt werden konnten, sind auf die Bronchiolen bis mittelgradigen Bronchien beschränkt, die sonstigen Partien sind fast frei davon.

**Morphologie der Acarina:** Körper grauweiss, 0,74—0,76 mm lang und 0,30—0,40 mm breit (Männchen kleiner als das Weibchen); breit gerundet nach hinten und mit ungegliedertem, mit dem Vorderleibe verschmolzenem Abdomen. Die Haut ohne sichtbaren Schild, mit Ausnahme der Stigmaplatte lederartig und zart, Palpen mit drei Gliedern und ganz zurückgezogen. Vier Paar Beine an der Unterseite des Vorderkörpers ohne chitinige Stützleisten (Epimeren); die Beine relativ kurz (0,27 mm) und kräftig, von fast gleicher Länge und in gleichen Abständen angesetzt, die zwei vorderen Beine etwas näher zusammengedrückt und aus sechs Gliedern bestehend. Das Endglied endigt in zwei gebogene, auseinander weichende Krallen, und darunter befindet sich ein breiter Haftlappen. Alle Glieder, ausgenommen die basalen, haben kleine Borsten. Die Stigmaplatte queroval, etwas chitiniert und beiderseits zwischen den Coxen des 3. und 4. Beinpaars liegend. Die Afteröffnung liegt an der Unterseite des Hinterkörpers



und ist auf beiden Seiten und vorn mit einer Borste versehen. An der Bauchfläche zwischen den hinteren Coxen befindet sich beim Weibchen eine querliegende Furche (Vulva). Vorn auf der Rückenseite sind einige kleine Borsten. Der Darm besitzt drei (wahrscheinlich) paarige Blindsäckchen. Ovarien einiger Weibchen fast völlig ausgebildet und schon gut entwickelte Eier enthaltend. Die Parasiten unseres Falls waren ausschliesslich erwachsene oder junge Individuen; in der Zyste konnten weder durch uns noch durch die beiden vorgenannten Forscher Eier nachgewiesen werden; doch weicht der Befund der beiden letzteren insofern von dem unsrigen ab, als eine sechsbeinige Nymphe vorgefunden wurde.

Gestützt auf die obengegebene Charakteristik der Acarina stimmt diese mit der Beschreibung von N. Bank<sup>1)</sup> über eine Lungenacarina d. h. *Pneumonyssus simicola* Bank überein.

Durch die obigen Untersuchungen haben wir bewiesen, dass die Veränderungen, die von der Acarina hervorgerufen werden, nur in den relativ kleinen Bronchien oder Bronchiolen nachzuweisen sind. Das Vorkommen der Acarina ist daher auf die obenerwähnten Bronchien oder Bronchiolen beider Lungen beschränkt; die Veränderungen, die der Parasit hervorruft, haben den Charakter einer chronisch verlaufenden reaktiven Entzündung, die an der Stelle, wo der Parasit schmarotzt, lokalisiert bleibt. Der Körper des fest an der Wand haftenden Parasiten ist stets ganz intakt. Durch diese Gründe erscheint es klar, dass die Acarina nicht erst nach dem Tode des Wirtes in die Lungen gelangt sein kann; aber auch die Möglichkeit der Einwanderung einer auf dem Leib schmarotzenden Acarina muss als ausgeschlossen angesehen werden; die Sache liegt wohl zweifellos so, dass der Parasit sich schon seit längerer Zeit in der Lunge eingenistet hat. Weiter hat er kein Auge; alle hornartigen Schilde mit Ausnahme der Stigmaplatte sind ganz zart; die Mundteile und die Palpe sind in den Kopf zurückgezogen und die Beine an ihrer Insertion weiter auseinander gerückt. Diese Eigenschaften weisen auf sein parasitäres Leben hin.

1) Bank, A new genus endoparas. Acar. (Geneesk. Tijdsch. v. Nedenl. Indiu. D. 41. 1901)

Zur weiteren Beleuchtung dieser Frage wollen wir mit gütiger Erlaubnis des Herrn Prof. Dr. K. YAMAGIWA<sup>1)</sup> aus dem Protokoll über eine am 11. Okt. 1895 ausgeführte Sektion eines an Lungendistomen verstorbenen Mannes, der an epileptischen Krampfanfällen gelitten hatte, einen Abschnitt citieren und mit unsern Ergebnissen vergleichen. Der pathologisch-anatomische Befund war wie folgt:

„Beide Lungen sind mit der Brustwand und dem Zwerchfell fest verwachsen. An der lateralen Seite des linken Unterlappens befindet sich eine indurierte Stelle, welche auf dem Durchschnitte sich als eine mit käsig-schmieriger Masse ausgefüllte Cyste zeigt. Bei genauer Untersuchung fand ich in dieser käsigen Masse ein schon gestorbenes, grau-dunkelbläuliches Exemplar von Lungendistomen (der lebende Wurm ist rötlich). Ferner wurde mikroskopisch nachgewiesen, dass in der Masse eine gross Anzahl von Eiern und Charcot'schen Krystallen enthalten war. Die Grösse des schon geschrumpften Distomum betrug 1 cm Länge, 1/3 cm Breite und 1 mm Dicke. Mund- und Bauchsaugnapf waren noch deutlich zu erkennen. Die Form der pflaumkerngrossen Cyste ist länglich abgeplattet, ihre Längsachse hat dieselbe Richtung, wie die der Länge, nur etwas schräg gestellt, d. h. der obere Teil liegt in der Adhäsionsschicht, der untere steckt im Lungenparenchym. Die Cyste hat eine einige Millimeter dicke Wandung, die sich nicht von dem sie umgebenden Gewebe abtrennen lässt. Die Innenfläche der Cysten-Wand ist gefaltet, rauh. Die Falten bergen oft Nebenträume zwischen sich. Dicht an der seitlichen unteren Wand der Cyste sieht man ektatische Bronchien. Eine freie Kommunikation, sei es von Gefässen, sei es von feinen Bronchien mit dem Cystenraum konnte ich nicht wahrnehmen. In dem Lungenparenchym, nahe an der Cyste, einige indurierte Partien. Sonst makroskopisch keine merkliche Veränderung im Ober- oder Unterlappen zu konstatieren. Im Adhäsionsgewebe hie und da hämorrhagische Herde. Die rechte Lunge zeigt keine Abweichung. In den Baueingeweiden und auch im Grosshirn habe ich keine für Lungendistomen sprechende Verände-

1) YAMAGIWA, Über die Lungendistomen-krankheit in Japan. (Archiv f. path. Anat. Bd. 127, Heft 3. S. 449, 1892)

rungen wahrgenommen. Vom obern Teil der Cyste mitsamt dem anliegenden Gewebe habe ich je ein Stückchen ausgeschnitten und mikroskopische Präparate daraus hergestellt. Die Untersuchung derselben hat Folgendes ergeben :

Die Cystenwand besteht aus einem zellenreichen Bindegewebe ; die dem Cystenraum zugekehrte Fläche schickt feine Fasernetze geben den Cystenraum hin. Blutkörperchen, pigmenthaltige Epitheloidzellen und Charcot'sche Krystalle liegen zwischen den Fasern und an der relativ glatten Innenfläche. Ferner sieht man Distomeneier hie und da in die Cystenwand eingebettet liegen. Unter diesen gibt es auch solche, welche Kapillaren oder Bronchiolen zu verstopfen scheinen. Um die bindegewebige Schicht sieht man starke Rundzellinfiltration, auch geschlängelte Gefässe. In der Wand, die an das Lungenparenchym grenzt, sind noch erhaltene Alveolen und Bronchiolen vorhanden, erstere mit pigmenthaltigen Epithelzellen gefüllt. Auch finden sich Distomeneier und Riesenzellen von unregelmässiger Gestalt (im Zentrum der Zelle viele Kerne, nicht in der Peripherie). Ferner Gebilde mit dem Aussehen von Riesenzellen, welche Distomeneier in sich fassen. Die Untersuchung der Schnitte auf Tuberkelbazillen ist negativ ausgefallen."

Aus diesen Ergebnissen der makroskopischen und mikroskopischen Untersuchungen zu urteilen, lässt sich eine grosse Ähnlichkeit mit unserem Falle nicht übersehen, obwohl leichte Abweichungen vorhanden sind, nämlich :

1. a) Die obenerwähnten zystischen Knötchen befinden sich dicht unter der Pleura und tief in dem Parenchym multiple in beiden Lungen.
- b) Ein zystisches Knötchen an der lateralen Seite des linken Unterlappens der Lunge.
2. a) Die Grösse der Zyste schwankt zwischen Erbsen- und Milliargrösse.
- b) Die Grösse der Zyste ist pflaumkerngross.
3. Bei beiden Fällen sind die Veränderungen der Zystenwand und deren Umgebung fast gleichartig, doch mit dem Unterschied, dass sie in

unsrem Falle geringer sind.

Die von uns konstatierten Veränderungen der Lunge stimmen mit den durch Lungendistomen verursachten Abweichungen überein. Obwohl nun der Lebenskreislauf des Parasiten unklar ist, und Eier in der Zyste nicht gefunden wurden, so ist es doch sicher, dass der Parasit in seinem erwachsenen Stadium ein endoparasitäres Leben zu führen vermag.

### **Schluss.**

1). Gestützt auf die Morphologie der Acarina unseres Falles lässt sich dieselbe als zur Art *Pneumonyssus simicora* Bank zugehörig bestimmen.

2). Diese Acarina schmarotzt nur in den Lungen, besonders in den mittelgradigen Bronchien und Bronchiolen.

3). Diese Bronchien und Bronchiolen werden durch den Reiz der Acarina hier und da zystisch erweitert, ihre Wände stark verdickt. Die Krankheit zeigt das Bild von Bronchitis et Peribronchitis catarrhalis chronica.

4). Die affizierte Lunge zeigt fast gleiche Veränderungen wie diejenigen, welche beim Menschen durch Lungendistomen hervorgerufen werden. Die Acarina haftet fest an den Wänden der Bronchien und Bronchiolen.

5). Berücksichtigen wir die morphologischen Merkmale und besonders die Beschaffenheiten der Haut und der Palpen zusammen mit den oben erwähnten Lungenveränderungen, so glauben wir uns zur Annahme berechtigt, dass der Parasit in seinem erwachsenen Stadium ein endoparasitäres Leben führen kann.

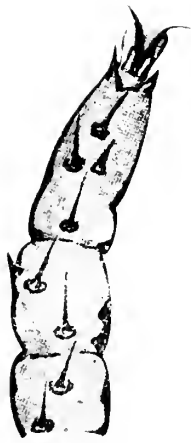


Fig. 2



Fig. 1



Fig. 3



Fig. 4

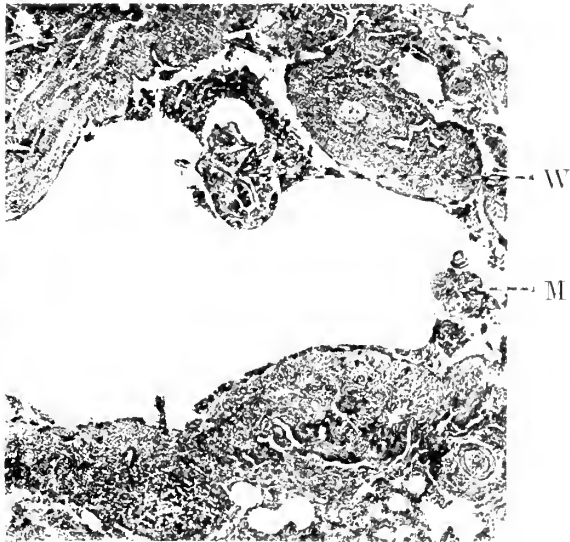


Fig. 5



### Erklärung der Tafel VI.

- Fig. 1. Kopfteil von *Pneumonyssus simicola* Bank (Dorsalansicht). Die punktierte Linie zeigt das vorderste Brustende. Vergr. ca 275.
- Fig. 2. Die Unterhälfte des ersten Beines. Vergr. ca 400.
- Fig. 3. Die Unterhälfte des vierten Beines. Vergr. ca 400.
- Fig. 4. Zwei horizontal und quer geschnittene Individuen von *Pneumonyssus simicola* Bank in einem tief in dem Lungenparenchym liegenden kleinen Bronchialast. Färbung : Hämatoxylin-Eosin. Vergr. 30 : 1. o Ovarium ; v Verdauungstraktus ; M Männchen ; W Weibchen.
- Fig. 5. Zwei horizontal und quergeschnittene Individuen von *Pneumonyssus simicola* Bank in einem dicht unter der Pleura liegenden zystisch erweiterten Bronchiolus. Färbung : Hämatoxylin-Eosin. Vergr. 30 : 1. M Männchen ; W Weibchen.

# ON THE INHERITANCE OF AN AURAL ABNORMALITY IN THE AYRSHIRE CATTLE.

By

**Jinshin Yamane,** *Nōgakushi.*

---

The present paper deals with genetical studies upon a case of abnormality, which has been transmitted with striking persistence for more than half a century.

In the Ayrshire cattle a very singular abnormal peculiarity is frequently seen, which is known amongst breeders as nicked or notched ears.<sup>1)</sup> This aural abnormality is not a traumatical but a natural one, it always occurs symmetrically in both ears. In the size of the notch, however, there are great differences among the affected individuals, and two distinct types can be distinguished. In some animals the tips of the ears are slightly notched : whilst in others it will be observed that the notches are so conspicuous that the ears seem to be not more than half the usual size. In the latter case the ridges on the inner surface of the lobe are exceedingly developed. The degree of affection is entirely constant at all ages. Rough sketches below indicate particular features, where a verbal description can not adequately explain relative differences. For convenience' sake I will call the well-notched one the first type and the slightly-notched the second type.

It would be of deep interest to know the origin of this aural abnormality in this breed of cattle, but it is naturally veiled in obscurity. A slight suggestion, however, may be obtained from a letter that was sent in 1902 to Mr. S. Takenouchi, the director of the Mayeda Farm in Ishikari Province,

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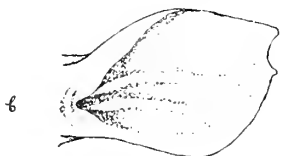
1) The terms "nicked" or "notched" are used in the American Ayrshire Record.

[Jour. of the College of Agr., Tohoku Imp. Univ., Sapporo, Vol. VI, Pt. 7, April, 1915]



and one of our prominent Ayrshire breeders, by Mr. G. Bement, Oakland Cal., U. S. A. A quotation from Mr. Bement's letter reads: "The nicked ears above are natural and can be traced back to the imported bull Eglinton

(21) described in the Herd Book as 'Dark Brown and White with Nicked Ears.' Many of my herd were calved with nicked ears."



Diagrams showing the normal and abnormal ears in the Ayrshire breed.  
a, Normal type; b, Slightly nicked type;  
c, Well nicked type.

According to my examination of the Ayrshire Record I and II<sup>1)</sup> five bulls and eight cows are recorded to have had such abnormal ears. With the single exception of the bull "Carleton Chief" (1568), all of these were the descendants of "Eglinton." No doubt there must be more affected individuals but many of them, I think, either have escaped notice, or have not been described as to their aural peculiarity. As the bull Eglinton was imported in 1859 from Scotland to the U. S. A., the remote origin of this abnormality should naturally be sought in its native habitat.

In our country this peculiarity of the ears can be traced almost without break to a bull "Express" (4503), once owned by the Mayeda Farm. At this farm he has produced more than 35 offspring, all of them having this characteristic. "Express" was bred in 1888 by Mr. G. Bement, above cited, imported in 1890 by Mr. M. Hori, and sold in 1894 to the Mayeda Farm. As he was a superior animal, his stock enjoyed great celebrity and popularity in its day and the nicked ears were highly valued as a distinctive indication of his

1) The Ayrshire Record I. 1876, Boston.  
The Ayrshire Record II. 1878, Boston.

blood. It is said by our farmers that the animals of the "Express" family were generally small, but excellent milkers and remarkable for their feeding qualities. Many years elapsed, however, before any signs of degeneration declared themselves; eventually the animals became smaller, less fecund and the strain lost its once appreciated qualities. Consequently, there are now very few animals retaining the "Express" blood at the Mayeda Farm, most of them having been eliminated.

Recently I paid visits to this farm and to a number of breeders who have once introduced the "Express" blood in their herds, and endeavoured to collect some accurate details on the inheritance of the aural abnormality.

The genealogical table accompanying this paper has been compiled either by studying the breeding books kept at the Mayeda Farm or by examining the living animals. All the cases, where there has been the least doubt as to the individual being normal or abnormal, have been excluded in the table. The normals are represented by white circles and the abnormals, whose type is unknown, by shaded circles. In the case of such abnormals, as I have been able to verify myself by actual examination of the individuals, I have preferred to distinguish the well-nicked type (the first type) and the slightly-nicked type (the second type) with black circles and black semi-circles respectively. In the table, the two lines coming together at the top of an individual trace respectively to the sire and dam, while the lines from the foot of the individual run to the offspring.

Owing to the lack of attention paid to this peculiarity on the part of the breeders, who regarded its inheritance as common rather than occasional, only a few records telling of this abnormality have come to my knowledge. As a matter of fact, the data thus far collected are decidedly fragmentary, but enough has been given to show the persistence with which the abnormality was transmitted.

In all cases cited in the table it must be admitted that the aural abnormality is inherited only from the abnormals and never from normals. An

abnormal sire No. 39 with a normal pure-bred Holstein-Friesian cow No. 41, whose origin could never be taken for heterozygous with respect to the aural peculiarity, has given an abnormal heifer No. 60. There are, therefore, indisputable evidences of dominance of this aural characteristic.

I have already mentioned that this peculiarity of the nicked ears is very different in degree. As far as my collection is concerned, sure cases of the first type were found in the bulls "Express", No. 25 and No. 39 and in the cows No. 61, No. 62, and No. 67. That the bull "Express" had well-nicked ears was stated to me by Mr. Takenouchi, who kept this bull for seven years. In five other animals it was ascertained by myself, as they are still alive at the present time. Further information from Mr. Takenouchi shows that all the offspring from "Express" were calved with nicked ears. This proves this bull to have been homozygous in regard to the aural abnormality. It appears that the other five animals, above mentioned, might also be homozygotes, considering that they descended from abnormal parents and moreover that no normal offspring from them has so far been detected. These data lead one to conclude that the well-nicked type is pure dominant (DD).

Turning to the second type, i. e. those which were calved with slightly nicked ears, we see in the genealogical table that all animals belonging to this type are the progeny coming from parents, one of whom is abnormal, the other normal. In other words, the individuals with slightly-nicked ears must be heterozygotes (DR).

It would seem, therefore, that the homozygous dominant is externally distinguishable from the heterozygous one.

We see further in the table that the abnormality is sometimes not transmitted (cows No. 35, No. 70 and No. 71). I have endeavoured to get some further details as to the numerical ratio between the abnormal and normals. The only data obtainable were the results from crossings between abnormal sires and normal dams at the Kanei Farm in Teshio province and at the Nemuro Farm in the province of Nemuro. Each farm now owns two bulls of the second type, all being descendants from "Express." In the past two years

at the former farm and last year at the latter farm, the following results were secured :

| Abnormal<br>sires <sup>1)</sup> | Number of<br>matings | Abnormal<br>offsprings | Bred at     |
|---------------------------------|----------------------|------------------------|-------------|
| No. I                           | 7                    | 5                      | Kanci Farm  |
| No. III                         | 16                   | 9                      | "           |
| No. 120                         | 6                    | 2                      | Nemuro Farm |
| No. 126                         | 3                    | 2                      | "           |
| Total                           | 32                   | 18                     |             |

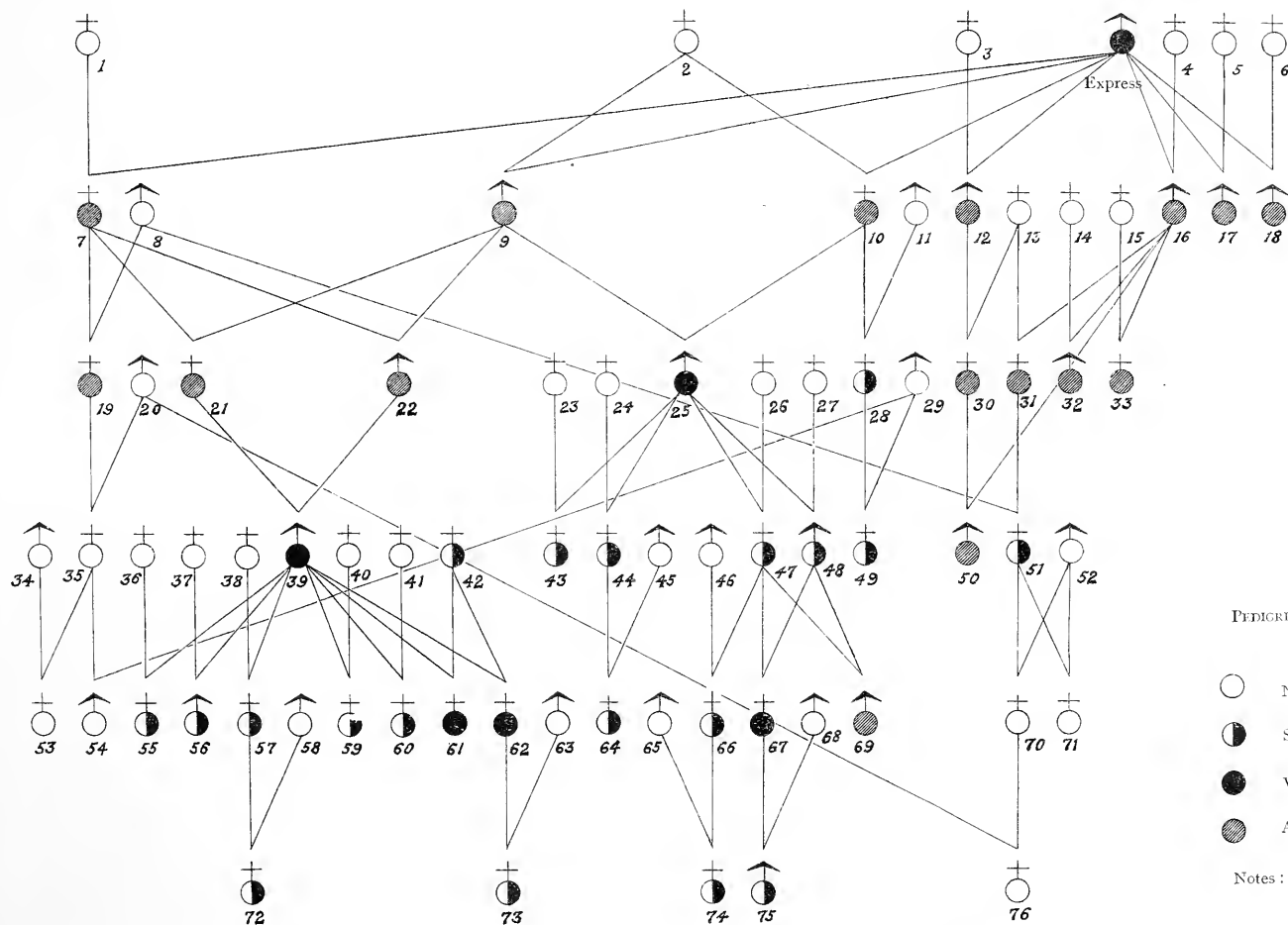
The calculation shows that of all the matings 56.3 % or nearly the theoretical 50 % belonged to the abnormal type. At all events the nicked character of the ears is either inherited or not inherited entirely. That is to say, there occurs a clear-cut segregation of the abnormality from the normality.

In recapitulation, it may be said that the data dealt with in this paper show that the nicked ears seen in some families of the Ayrshire breed originated at first in Scotland more than fifty years ago ; that this aural abnormality is transmitted in full accord with Mendelian principles ; that the zygotically different types DD and DR seem to be externally distinguishable in this case.

I wish here to express my heartiest thanks to Prof. S. Hashimoto and Assistant-Professor Y. Tanaka, to whom I am indebted for valuable suggestions in completing this investigation. I am also under obligation to Mr. K. Takenouchi and many other breeders for their kindness in placing their herds and breeding-records at my disposal.

From the Zootechnical Institute,  
College of Agriculture,  
Tohoku Imperial University.

1) These sires are numbered in accordance with the breeding records of both farms.



PEDIGREE ILLUSTRATING DESCENT OF THE NICKED EARS IN THE AYRSHIRE BREED.

- Normal type
- ◐ Slightly nicked type
- Well nicked type
- ◑ Abnormal, but the type unknown

Notes: No. 41, a pure-bred Holstein-Friesian Cow.  
No. 42, a descendant from "Express", but its continuous ancestors unknown.



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APR 29 1924

## ON THE ERGOT OF EQUIDAE.

By

**Schin. Yoshida.**

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In 1913 I wrote the work entitled "Morphologische und Physiologische Bedeutung der Sogenannten Kastanie an den Gliedmassen der Equiden", for the Zoological Institute at Halle a. S. In this work, the outcome of a great deal of microscopic and macroscopic study, I believe I have given a definite answer to the question "What is a Callosity?" but I did not then give any results of histological research on the Ergot, a small hornified mass found in the tuft of hair at the back of the fetlock. Since that time I have made further histological observations, the results of which I give below.

In order to do this I wish first to refer to the results of former zoologists.

Flower, in "The Horse", says, "The Ergot in the horse corresponds to the Afterclaw in the tapir, which is of the greatest use to the animal".

In 1903, Lydekker, in "Proceedings of the General Meetings for Scientific Business of the Zoological Society of London", Vol. 1 wrote a great deal on the subject of Callosities without mentioning the Ergot. Against Ewart's theory, published in "The Royal Society of Edinburgh", that the Callosity is a rudimentary toe, he wrote as follows: "A more important objection to the foot-rest theory is to be drawn from the fact that the fore Callosities are above the Carpus and are, therefore, too high to serve as foot-rests to any plantigrades, and as it is impossible that any change of position should have taken place, they never could have served as foot-rests."

The hind Callosities lie, on the contrary, below the hock and are therefore on a part of the leg which, by plantigrades is considered the foot surface; but while the fore Callosities are altogether homologous with the hind, it is

clear that as the fore Callosities are not foot-rests neither can the hind ones be.

If the Callosities are toe-rests, the ancestors of these families must, at the time when they were still plantigrades, have possessed them, but, so far as I know, no Ungulata was ever a sole-walker with all its limbs.

Lydekker has studied the Tarsal glands of reindeer and said that these corresponded with the Callosities of the horse and that the Ergots are rudimentary toes.

Ludwig Frank, in "Handbuch der Anatomie der Haustiere", second edition, page 796, has written on the microscopic structure of the Ergot and Callosity, that these form rudimentary horntubes and laminae.

Ellenberger, Stoss, Bonnet and many histologists have stated in their handbooks that the Ergot is of the same histological structure as the Callosity, that it is possible that the Ergot may be a rudimentary digit but that this has not been proved.

In 1913, when I wrote "Morphologische und Physiologische Bedeutung der Sogenannten Kastanie an den Gliedmassen den Equiden", my research into the matter of the Callosity had been thorough but I had made little histological observation on the Ergot. I had only made a macroscopical observation of the position and said that it might be a rudimentary pad.

In 1914, Zietzmann wrote in "Morphologie, Genese und Bedeutung von Kastanie und Sporn der Equidae", that from many deep microscopic, macroscopic and embryological researches he had concluded that the Callosity and Ergot are rudimentary pads.

Since that time, to determine whether the Ergot, like the Callosity, is a rudimentary digit or, on the other hand, a rudimentary pad, I have made further investigation which I shall now describe in the following order:

- 1.) Microscopic and Macroscopic Observations of the Ergot.
- 2.) Histological comparison of the Ergot and Pad.
- 3.) My own opinions on the subject.

I obtained material for observation from the slaughterhouse of Halle a.S by the kind favour of Professor Haeckel; first, fresh material preserved in

70% alcohol for three hours, after that in pure alcohol for twenty-six hours, in Xylol for twenty-four hours, and so on, then embedded in paraffin, stained with Vangieson's solution and Haematoxylin. The pad of the dog and cat I have preserved in the same manner.

I must express my gratitude to Professor Hacckel and his assistant Fraeulein Dr. Kuttner for the kind encouragement and advice they have always given me and also to Professor Thomas in the British Museum for his kindness in allowing me to inspect so much material.

### 1. Microscopic and Macroscopic Observation of the Ergot

The Ergot is the name applied to the mass of wart-like horn situated in the tuft of hair on the inner side of the fetlock. Its size varies in the case of different horses and with different ages. Among the Equidae, when the Ergot appears on both fore and hind feet, according to my measurements, it is larger on the latter than on the former, but in many cases it is found on the hind feet only, with no trace of one on the fore feet.

Generally, the Ergot is larger among the heavier breeds of horses than in the smaller breeds, but there are exceptions. Very small horses, as for instance native breeds, sometimes have a large Ergot on the hind foot, and in the case of English thoroughbreds it is sometimes larger than among heavy horses. In conclusion I will repeat that :

- 1.) When the Ergot is found on fore and hind feet it is larger on the latter than on the former,
- 2.) Sometimes it is found on the hind feet alone.

Professor Zietzmann's measurements of the diameter of the Ergot on the donkey are, on the forefoot, 2.0 cm., on the hind foot 2.3 cm. Its thickness, however, though small in comparison with the Callosity yet measures some millimeters.

Among Zebras, the diameter of the Ergot on the hindfoot is 2.0 cm. larger than that on the fore foot.

In the British Museum I have observed the following different equine breeds :

*Equus grevyi* (?). Ergot on fore and hind feet, very large on hind feet.

*Equus samalicus*. Ergot on fore feet  $1\frac{1}{2}$  mm. broad,  $2\frac{1}{2}$  mm. long, on hind feet,  $2\frac{1}{2}$  mm. broad, 3 mm. long.

Burchell's Zebra (*Eq. burchelli selousi*). Ergot on fore foot smaller than on hind foot.

Grey's Zebra. (*Eq. grevyi*). Ergot on fore and hind foot, that on hind very large.

As these specimens are stuffed the skin has shrunk somewhat causing the Callosity and Ergot to become smaller, but as I only desire to compare the relative sizes of fore and hind Callosities and Ergots this shrinking is of no account. Professor Zietzmann's research in the case of the Ergot has been very thorough. He says that among hinnies the Ergot grows as with horses to a considerable thickness (over 2 cm.), that those on the hind feet have the larger surface, the greatest diameter of which sometimes exceeds 2 cm. In the case of mules he had made no observations.

As I have said above, from the microscopic studies, the Ergot, in contradistinction to the Callosity which is of the same size on fore and hind feet, is always larger on the hind feet.

In my conclusion I wish to give an explanation of this.

In the first place, to examine the theory of many histologists that the Ergot is a rudimentary pad, I have compared it with the toe pad and central pad of dogs and cats, and in particular with the carpal pad which, at the present time, is not used by its possessor.

If the Ergot is a rudimentary pad it was formerly used by its possessor as a pad, and in my opinion its histological structure should be similar to, or the same as, that of the pads of dogs and cats. I have, therefore, first made histological studies of the pads of domestic cats and dogs as follows :

A.) Histological structure of the pads of cat and dog,

a) Pad of domestic cat.

The Stratum Mortificatum is composed of quite dry, flat, scaly cells, from which hornified scales are thrown off from the superficial layer. In the pad of the cat this layer is more compact than in the Chestnut of the horse.

The Stratum Mortificatum gradually merges into the Stratum Corneum with no distinct connecting line. This latter Stratum consists of layers of cells with hornified membrane, and large and small scales which are continually forming the Stratum Mortificatum. These are the dead elements. In this layer, the Marklayer of the supra papillar epidermis, which is to be seen in the epidermis of the Ergot, cannot be found.

Stratum Corneum is connected with Stratum Granulosum by Stratum Lucidum. This latter consists of hornified cells without nucleus; the cell wall is hardly to be seen even in thin sections, and is generally quite indistinct.

Stratum Granulosum is a quite thin layer, and is only to be distinguished from its upper layer, Stratum Lucidum, by staining with Hematoxylin as the nucleus of the cell of Stratum Granulosum is easy to stain. The cells of Stratum Granulosum are quite flat in form and contain a small round nucleus.

Stratum Spinosum or Stratum Malpighii in the pad of the cat is very well developed, and contains the cone-shaped Papillae which do not penetrate this layer as in the case of the Ergot.

The Prickle Cell of Stratum Spinosum is larger than in the Callosity and Ergot, and more distinct. The cells are joined by means of fibrils which pass from one to the other as in the case of other hornified cells.

Stratum Germinativum consists of cylindrical cells, containing a nucleus, and arranged in rows in quite thin layers which gradually merge into Stratum Spinosum with no distinct dividing line. When the pad is black this Stratum Germinativum contains much pigment, and when it is stained with Hematoxin takes a strong colour which distinguishes it from the other layer.

From this layer (Stratum Germinativum) the cells are continually being divided and driven upwards with the groups of pigment bodies and gradually

changed to Prickle Cells.

The Papillae of the pad of the cat are comparatively less developed and are cone-shaped. These are filled with connective tissue, which is joined perpendicularly to the tissue in the Stratum Reticulare. This latter tissue is finer and contains many small blood vessels.

In the Cutis Vera of these balls, hair roots and sweat glands are not found, but on the border line between the common skin and the pad many hair roots and sweat glands are found.

My histological studies of the pad of the cat have enabled me to verify the following important facts :

The Papillae of the pad of the cat are cone-shaped with broad base and pointed top, and they are enveloped with a layer of Prickle Cells. They never penetrate the Stratum Spinosum and keep a simple form.

b) Pad of the dog.

The histological structure of the pad of the dog differs very slightly from that of the cat. I have, therefore, to avoid repeating myself, described only the differences, which are as follows :

Stratum Corneum is more developed than in the cat and I have obtained the following average measurement : 0.45 mm.

Stratum Lucidum, as in the cat, is a very thin layer, fine and clear and difficult to stain.

Stratum Spinosum is a very simple wave-like layer and thin as in the cat. The Prickle Cells are difficult to stain and contain a distinct round nucleus, which, as the cells gradually reach Stratum Germinativum, becomes easier to color, and the cells themselves become flatter and flatter.

Stratum Germinativum, in the case of a colored pad, contains very many pigment cells in its lower part. This pigment is found in the bottom layer of cells which make a mitotic division, and it passes upwards with the divided cell.

The Papillae in this pad are larger than in the cat and are of an elongated cone form. They are enveloped by Stratum Malpighii, but their tops do not



penetrate Stratum Malpighii as they do in the Callosity and Ergot.

The histological structure of the pad of the dog is in general similar to that of the cat.

### **B. Histological Structure of the Ergot**

As I have related above, the Ergot, like the Callosity and the Pad, was originally a variety of the common skin, and its histological structure, therefore, should resemble that of the other two; when, however, it is studied in very minute detail some differences in the degree of divergence from the original are discerned.

The result of my histological study is as follows, starting from the surface layer downwards:

Stratum Mortificatum, in the Ergot, is very well developed and is very long. It consists of large and small scales, which, being delicate and flat, adhere closely together and are not easily rubbed off as in the pad of the dog and cat, and therefore grow long like a nail. As the surface of an undeveloped Ergot is comparatively flat, the thickness of Stratum Mortificatum varies with the degree of development, but the other layers remain unchanged.

Stratum Corneum, as in the Callosity and Pad consists of a layer of cells with hornified membrane, connected with a layer of Prickle Cells by Stratum Lucidum. In this layer Horntubes are to be found which are not seen in the Callosity and the Pad. These are formed by the tops of the elongated Papillae which penetrate Stratum Lucidum and the Prickle Cell layer. In a horizontal section Rohrwand cells and Horntube mark cells may distinctly be seen in these Horn tubes.

Stratum Lucidum, in the case of the Ergot, shows no distinct connecting line with Stratum Granulosum, and the former is to be distinguished from the latter only because it is more difficult to stain.

The Prickle Cell layer is very thick, with an average thickness of 1-3 mm. It is penetrated by elongated and well developed Papillae and divided by them into interpapillary and superpapillary epidermis. On the base of

the superpapillary epidermis many pigment groups are to be found.

Stratum Reticulare, which forms the Corium, contains fine fibres which always run parallel with the layer and perpendicularly to the papillary body. In these fibres blood and lymph vessels are embedded. In the case of the pad many fat cells are found in this tissue, but in the Ergot they are found, not in these fibres but in the Subcutis.

The fine elastic fibres in Stratum Reticulare, running into the Papillae with blood and lymph vessels, compose the so-called Stratum Corpus Papillary. When this is stained with Vangieson's solution these fibres can easily be distinguished from the Rinden surface of Stratum Germinativum.

The Rinden surface consists of one-layered cells, of somewhat cylindrical shape, containing a large nucleus which can be easily stained with Kaematoxin. The histological form of these cells is similar to those in the base of the Epithel of the Oesophagus, but the contents of the former cells are clearer than of the latter.

The Papillae of the Ergot, according to my measurements, are 1-2.9 mm. long; their number is greater than in the Chestnut and Pad and they are better developed. Their histological form is similar to that of the Papillae and Horn-tubes which are found in the claw wall of cattle.

In particular the Horntube Mark and Horntube Wall cells and the easily stained granulated cells, which are found in the hoof of the young horse, are also seen in the Ergot. I cannot find such well developed Horntube Mark and Horntube Wall cells in the Callosity and the Pad.

In my histological study of the Ergot I have remarked the following important points:

- 1) A well developed Papillae which penetrates the Prickle Cell layer, made up of the Horntube Mark in Stratum Cornium,
- 2) The number of Papillae is greater than in the Callosity and the Pad,
- 3) In all other respects it is similar to the Callosity.

As I have written above, from my microscopic studies, the histological structure of the Ergot is the same as that of the Callosity, especially in the

full development of the Papillae. Such well-developed Papillae are not found in the Pad and other common skins. That the histological structure of the Ergot resembles that of the Callosity has already been declared by Professor Ellenberger and other zoologists. Professor Zietzschmann and others have said that the Ergot and the Callosity are rudimentary pads. If this were the case, their histological structure should be the same as, or at least be similar to, that of the Pad. This resemblance I have not been able to find in my studies, and therefore believe that the Ergot and the Callosity are rudimentary digits.

That, by means of histological and embryological studies, I find the Callosity to be a rudimentary digit, I have already explained in "Archiv fuer wissenschaftliche und praktische Tierheilkunde", Band 39, Heft 6, 1903; from which I will only repeat the most important points for the sake of clearness. At that time I proved my opinion from Papillae in all grades of development. These I have divided into four periods:

- 1) In this stage the Papillae have a flat outline, very little waved, and are found in less developed skin as the hornified tissues in a very early stage.
- 2) In this stage the Papillae are cone-shaped. These are found in the sole pad of the cat and dog.
- 3) In this stage the Papillae penetrate other layers and reach the hornified surface. These are found in the Horn of a three month's calf and in the Callosity of a full grown horse.
- 4) In this stage, fully developed and secondary Papillae are found, of very complicated form, as can be seen in the Hoof and Claw.

Thus the hornified tissue can be distinguished according to the grade of development of the Papillae. Now let us see to which grades the Papillae of the Callosity, Ergot and Pad belong.

As I have written in 1913, the Papillae of the Callosity belong to the third grade of development. After still further research I am in a position to declare that the Papillae of the Ergot belong to the third grade and those of the Pad to the second grade of development.

And now I should like to ask the theorists who affirm that the Callosity and Ergot are rudimentary Pads the following question. "Why are the Papillae and hornified conditions of the hornified organs, the Callosity and Ergot of the horse, which to-day are useless to their possessors, more developed than that of the pad of the cat and dog which is still a useful organ to its possessor.?"

If the Callosity and Ergot are rudimentary pads, the histological structure of the Papillae should be the same as, or similar to, that of the pad.

The rudimentary pad theorist might perhaps say that the pad of the horse had been in a more developed state than that of the dog and cat, but when the histological structure of the Frog is observed the Papillae are found to be less developed than in the Callosity and Ergot and the tops do not penetrate the Prickle Cell layer but are merely enveloped by it, precisely similar to that of the pad of the dog and cat.

From this I infer that the Ergot and Callosity were formerly digits and that the Frog of the hoof is a rudimentary ball.

Professor Zietzmann in the "Morphologie, Genese und Bedeutung von Kastanie und Sporn der Equiden", published his opinion that the Callosities and Ergots of Equidae are rudimentary pads, thus supporting Ewart's and Hinntze's theory. His histological and embryological investigations were very extensive and in opposition to the rudimentary toe theorists he wrote (page 12):

"The histological structure of hoof, claw and nail have quite another characteristic, and even the peculiarity that a prominent mass (hoof, claw, nail) composed of horn cells and closely bound to the underlayer is thrown off from the place of its origin over a wide surface of the cutis vera before it appears with its distal edge. This characteristic procedure affects such organs in the same manner as a stamp, heavily pressed would, so that it appears quite distinctly in the most rudimentary stages. I am thinking especially of the hind claws of ruminants and particularly of some kinds of deer, which are very little developed but which clearly show the aforesaid growth ten-

dency. Nothing of this, however, is to be seen on either the Ergot or the Callosity."

The question may be put by other theorists: "Why, if the Callosity and Ergot are rudimentary digits, are they found in such impossible positions?"

Before answering this question, I should like to give the results of my observations of the Callosity and the Ergot. I have already published the greater number of these observations of the Callosity in 1913. Since that time, however, I have made further studies of the Callosities of several other species of Equidae and of the Ergots of all Equidae.

| Race name                                                       |       | Fore Callosity |         |        | Hind Callosity |         |        |
|-----------------------------------------------------------------|-------|----------------|---------|--------|----------------|---------|--------|
|                                                                 |       | Length         | Breadth | Height | Length         | Breadth | Height |
|                                                                 |       | cm.            | cm.     | cm.    | cm.            | cm.     | cm.    |
| Belgian                                                         | left  | 3,6            | 5,6     | 6,23   | 2,16           | 3,7     | 4,66   |
|                                                                 | right | 3,4            | 5,46    | 6,00   | 2,33           | 3,65    | 4,51   |
| Persian                                                         | left  | 3,82           | 4,7     | 6,4    | 2,1            | 4,2     | 4,1    |
|                                                                 | right | 3,62           | 4,6     | 6,4    | 2,1            | 4,1     | 4,5    |
| Clydesdale                                                      | left  | 3,1            | 4,0     | 4,05   | 2,4            | 3,6     | 2,6    |
|                                                                 | right | 3,1            | 3,7     | 3,8    | 2,4            | 1,65    | 3,05   |
| English Thoroughbred                                            | left  | 1,4            | 2,9     | 0,7    | 0,87           | 1,5     | 0,55   |
|                                                                 | right | 1,45           | 2,98    | 0,75   | 0,75           | 1,55    | 0,55   |
| American Trotter                                                | left  | 2,0            | 3,9     | 0,76   | 1,5            | 2,4     | 0,66   |
|                                                                 | right | 2,0            | 3,8     | 0,8    | 1,4            | 2,2     | 0,6    |
| English Thoroughbred, reared in Australia.                      | left  | 2,0            | 4,0     | 0,8    | 1,7            | 2,3     | 0,5    |
|                                                                 | right | 2,0            | 3,8     | 0,6    | 1,7            | 2,3     | 0,4    |
| Japanese horse × English Thoroughbred (½ English Thoroughbred.) | left  | 2,3            | 4,1     | 0,7    | 1,5            | 2,0     | 0,46   |
|                                                                 | right | 2,5            | 4,2     | 0,6    | 1,5            | 2,0     | 0,43   |

As the above table shows, the Callosities of the heavy horses are larger and more fully developed than those of the lighter breeds, but in both cases they are larger on the fore than on the hind feet.

With donkeys and zebras it is only to be found on the fore feet. By examination of the stuffed specimens of different breeds in the British Museum, I have obtained the following results:

| Race Name                                             |          | Fore Callosity |            |        | Hind Callosity |          |          |
|-------------------------------------------------------|----------|----------------|------------|--------|----------------|----------|----------|
|                                                       |          | Length         | Breadth    | Height | Length         | Breadth  | Height   |
| Equus Grevyi                                          | (a) left | cm.<br>,6      | cm.<br>1,0 | —      | cm.<br>—       | cm.<br>— | cm.<br>— |
|                                                       | right    | ,8             | 1,7        | —      | —              | —        | —        |
|                                                       | (b) left | ,9             | 2,1        | —      | —              | —        | —        |
|                                                       | right    | 1,1            | 2,0        | —      | —              | —        | —        |
| Equus Samalicus                                       | left     | 2,0            | 4,0        | —      | —              | —        | —        |
|                                                       | right    | 2,1            | 4,3        | —      | —              | —        | —        |
| Equus Burchelli<br>(Burchelli Zebra)                  | left     | 2,0            | 3,2        | —      | —              | —        | —        |
|                                                       | right    | 2,1            | 3,4        | —      | —              | —        | —        |
| Equus Asinus<br>Africanus (Nubian<br>Wild Ass)        | left     | 2,3            | 2,4        | —      | —              | —        | —        |
|                                                       | right    | 2,2            | 2,5        | —      | —              | —        | —        |
| Equus Caballus<br>Prezwalskii (Mongolian<br>Wild Ass) | left     | 1,5            | 3,0        | —      | 0,3            | 2,2      | —        |
|                                                       | right    | 1,6            | 2,9        | —      | 0,4            | 2,1      | —        |
| Equus Heminous<br>Kiang                               | left     | 4,1            | 7,4        | —      | —              | —        | —        |
|                                                       | right    | 4,0            | 7,0        | —      | —              | —        | —        |
| Equus Quagga                                          | left     | 2,9            | 5,0        | —      | —              | —        | —        |
|                                                       | right    | 2,8            | 5,3        | —      | —              | —        | —        |

In the case of the Callosities of the bastards, Professor Ewart has published his observations in the "Proceedings of the Royal Society of Edinburgh", p. 467. He writes, "Among the Esquimaux hybrids the Callosity is absent; among Mongolian hybrids it is absent on the right fore foot and quite small on the left; while on the zebra hybrids it is very large on the hind feet." Many other zoologists have also recorded the results of their studies of the Callosities of the mule, eselzebroid and hinny. All these observations point to the same conclusion that when Callosities are found on both fore and hind feet, they are larger on the former than on the latter; and that when only one pair is found, this is always on the fore feet. On the contrary, when Ergots are found on all four feet, they are larger on the hind feet than on the fore; and when only one pair is found, this is always situated on the hind feet.

Thus we see that while the histological structure of the Callosity and Ergot is similar, the position of the larger pair is in the one case on the fore feet and in the other case on the hind feet.

How is it that such a peculiarity should have arisen among Equidae?

To account for it I will first write of the Callosity. That the Callosity is a rudimentary nail from its histological structures, and the reason of its being found either on the fore feet only or when on both fore and hind feet larger on the former than on the latter, I have already explained, but will now briefly repeat.

When the digits of different Mammalia are observed, it is found with a few exceptions that when the number on the fore and hind limbs is different, this is always greater on the fore than on the hind limbs, and that when fore and hind limbs have the same number, those on the fore limbs are always more developed than those on the hind.

Man and the man-like apes have five perfect digits on all four extremities, but those on the hind limbs are smaller than those on the fore.

Among dogs, five toes are found on the fore legs and four only on the hind legs, sometimes with a fully developed fifth.

Cattle have four toes on the fore and hind feet, with two degenerated so-called after-claws.

Tapirs have four toes on the fore feet and three on the hind feet.

Parameles have three toes on the fore feet with rudimentary first and fifth, on the hind feet two toes with rudimentary first, second and third.

The Equidae have one toe on both fore and hind legs.

Why is there this difference between the digits of the fore and hind limbs?

The fingers of the fore extremities serve to take up nourishment and for purposes of suckling, while those of the hind extremities are only used to support the body or for movement, and therefore they need to be more developed on the fore than on the hind limbs.

When we observe the many specimens in the Natural History Department of the British Museum, and if we believe the theory of English naturalists of the development of the Equidae, we can understand that the Callosity, which from its histological structure is a rudimentary digit, is always more

fully developed on the fore limbs. For example, *Hyracotherium*, found in the lower Eocene, had four fully developed toes on the fore feet and two functional lateral toes on the hind feet; other *Paracotherinae* in the Eocene and *Oligocaen* had somewhat rudimented lateral toes further from the ground, so that the function was lost, and the present horse uses only one toe to walk with.

This theory has been proved by many naturalists, so that, from the development of the horse, we see that the fore foot has always borne a larger number of fully developed digits than the hind foot.

If we observe this development of the digits of the horse, and the condition of the digits of all present mammals, that the greater development is always found on the fore feet, also that the callosity is either found on the fore feet alone, or is larger on the fore than on the hind feet, we must conclude that these facts are connected.

From my own histological observations and from these three facts, I infer that the Callosity must be a rudimentary digit.

For the theorists who affirm that the Callosity is a rudimentary pad, I must write that with the ox and deer which are now true digitigrades, the organ that corresponds to the pad is greatly rudimented and is sometimes scarcely to be seen, especially the pad which should belong to the after-claws. Only at the junction of the claw wall and the common skin can a slight trace be discerned, but sometimes not even that.

That, among the present Equidæ, whose remote ancestors—those at least earlier than the *Hipparion*, as fossils indubitably show—had already developed from plantigrades to digitigrades—a large rudimentary pad should still be seen, is quite impossible!

In order to compare the after claw with the ergot I should like first to write something of the after claw.

The after claw of Ungulata, which is found on the fetlock,—of what use is this to its possessor? It is clear that this organ, which is not so rudimented, that is, not so far from the ground, as in the ox and deer, is found



among animals so that when they walk on soft or marshy ground they may not sink too deeply into the yielding earth, and when the animal is accustomed to walk on steep hilly ground the after claw is needed as a support. These less rudimented after claws of the ox and deer are of similar development on fore and hind feet, but among the Ungulata, which have a quite rudimented after claw, we find a difference in the size on the fore and hind limbs. In particular, the after claw on the forefoot of the Madoquo Phillipsi is very small and that on the hind foot very large.

I have obtained the following result, that the more rudimented and useless the after claws the nearer they are together. For example, the fully rudimented after claws of the *Dorcotragus Megarotis* (Pl. VIII. Fig. 2), especially on the forefoot are quite close together and appear as one; the claw wall of its after claw has been thrown off and it has the appearance of a pad.

In particular, when the histological structure of such a fully rudimented after claw is seen, the Papillae are simpler than in the Ergot and Callosity of Equidae, and the hornified cell layer is thicker, that is the Stratum Corneum is not degenerated, than in the Ergot and Callosity.

That the Callosity and Ergot which have a more developed histological structure than the after claw of the *Dorcotragus*, should be rudimentary pads is not to be considered, because, as I have written above, the pad which is still functional is less developed than the Callosity and Ergot. I must here describe what I mean by the development of a hornified mass, which I have sometimes mentioned. This is the degree of development of the Papillae in the hornified tissue and of the horny layer. Because the Papillae always contain fine blood vessels which carry nourishment to the layers above, when the Papillae are developed the layers above will be equally so; such a fact may be seen in every hornified mass found on mammals, for example, in the fully developed hoof of the horse quite complicated Papillae may be seen with secondary Papillae. From such complicated Papillae to those of simple, nearly flat form which are found in the tissue of the very little developed hornified skin, the degree of development of the hornified mass is divided

into four classes as I have already described.

In the hoof, however, at the early embryological stage, the Papillae are quite simple.

The above results of my own observations show that the Callosity and Ergot which the present Equidae carry must be rudimentary toes.

I now wish to write of the frog, which, found in the hind part of the hoof, of Equidae, is a rudimentary pad which belongs to the toe, in order to ascertain its morphological connection with the Callosity and Ergot.

I have measured the frogs of various kinds of Equidae with the following results :

| Race Name                               | Fore Hoof |        |       |       | Hind Hoof |       |       |       |
|-----------------------------------------|-----------|--------|-------|-------|-----------|-------|-------|-------|
|                                         | a. b.     | c. d.  | e. f. | g. h. | a. b.     | c. d. | e. f. | g. h. |
| <i>Equus Burchelli</i>                  | 5½ cm     | 11 cm  | 6,2cm | 1½ cm | 7½ cm     | 10 cm | 6½ cm | 2 cm  |
| <i>Equus Grevyi</i>                     | 3,2cm     | 8,2cm  | 6,2cm | 1,5cm | 4 cm      | 8 cm  | 7 cm  | 2 cm  |
| <i>Equus Chapnani</i><br>(a) left side  | 6½ cm     | 11½cm  | 6 cm  | 2½ cm | 7 cm      | 10 cm | 6 cm  | 3 cm  |
| <i>Equus Chapnani</i><br>(b) right side | 6 cm      | 10,2cm | 6,4cm | 1,4cm | 5,8cm     | 10 cm | 7 cm  | 2 cm  |

a. b.....distance of two bases of bulbar expansion of periopilic ring, which is seen to be continuous with the sensitive frog

c. d.....distance between middle point of frog base and point of hoof

e. f..... length of frog

g. h.....breadth of frog

As these show, with most Equidae, the frog of the hind foot is always larger than that of the fore foot, especially in the Kiang and Grevy's zebra, where the frog of the hind foot touches the ground. It is clear that this has come from the fact that the angle which the hind foot makes with the ground is always smaller than that of the fore foot. We can tell that the present frog is still useful as a pad to the hoof. For the same reason we may infer that the Ergot formerly served as a support to the foot and was used at a later period by the hind foot than by the fore, and is therefore more rudimented on

the former than on the latter, also that the fore foot is a truer toe-walker with one hoof than the hind foot.

Finally, I have classified the Equidae according to the number and position of the Ergots and Callosities, which are rudimentary toes, in order to find out what condition the rudimented toe has on different kinds of Equidae and how the finger and toe have developed, as follows:

1. With Callosity and Ergot on both fore and hind feet.
2. With Callosity on fore foot but not on hind foot, and Ergot on hind and fore foot.
3. With Callosity on fore foot but not on hind foot, and Ergot on hind but not on fore foot.
4. No Callosities or Ergots on either fore or hind feet.

To compare with the Ungulata I have given the following table:

1. With two after claws on both fore and hind feet, all of the same size.
2. With two after claws on both fore and hind feet, but larger on the hind than on the fore feet.
3. With no after claws.

From the above table I can draw the following inference: from the fact that the fore extremities are always used for picking up food or for means of attack, their digits are more developed than those of the hind which are only used for support or to assist movement. The Callosity, which, as written above, is always more developed, must be the rudimented organ which was formerly used by Equidae for picking up food or as a weapon of attack. There must also be some connection between the facts that the Ergot is always larger on the hind foot among Equidae, and the after claw among cattle.

In order to make this clear, if the foot bones of those animals which now possess five fingers are compared with those of the Equidae, the following conclusion may be drawn: the first finger of Equidae was the earliest rudimented, as the hind foot finger of the present dog, then the second and fifth fingers were rudimented, which condition may be seen in the foot of the present pig. Still more rudimented second and fifth fingers are to be seen in

the after claws of cattle. This was the development in the case of the Ungulata, but with the Perisodactira the first finger was rudimented, then the fifth, fourth and second in order. That the fourth finger was rudimented earlier than the second may be seen from the splint bones of the present Equidae, because the inner splint bone is always larger and longer than the outer. From this fact I infer that the Callosity is the nail of the second toe and the Ergot of the fourth toe. But when the present position of the Callosity and Ergot is observed it appears to be a curious place for a nail to grow. To that I will answer that in a perfect finger the nail should be at the tip, but in the case of a rudimented finger there is no connection between the finger bone and the nail. A good example of this may be seen in the nail of the first finger of the hind foot of a dog, the position of which has so greatly altered. In particular, when the finger bone has no function as finger and the nail is still of use to its possessor, the latter always remains in a useful position, and the bone only is rudimented. From this fact, with the Equidae, as the second and fourth fingers only were rudimented, the nail of the fourth finger, the Ergot, was of use, like the after claw of cattle; the nail of the second finger of the foot remained as a means of attack, and it can be seen that when a horse fought with its enemy, hugging him with the fore feet, the Callosity might have been a useful weapon.

From the above morphological and histological results it is not to be believed that the Callosity and Ergot which present Equidae bear are, as Zietzmann and other zoologists state, rudimentary pads, and I must affirm that they are rudimentary digits.

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**Explanation of Plates****Plate VII.**

- Fig. 1. Cross section through Ergot  
 Fig. 2. Cross section through Pad of Dog  
 Fig. 3. Cross section through Pad of Cat

**Plate VIII.**

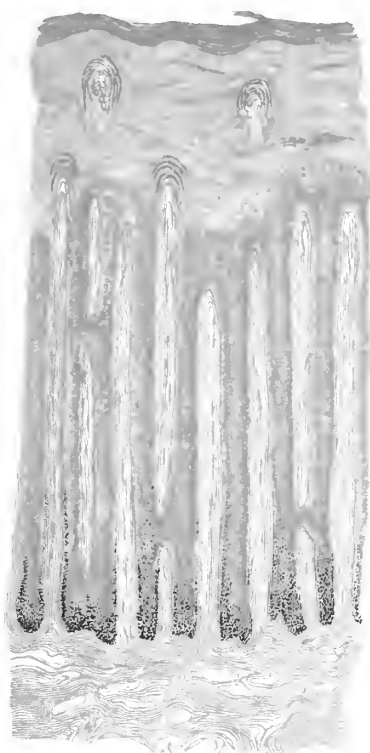
- Fig. 1. Cross section through Ergot. (original size)  
 Fig. 2. Fore and hind Foot of *Dorcotragus Megarotis*. (p—pad)  
 (1/3 size)  
 Fig. 3. Diagramatic view of fore and hind Hoof of Horse. (a—fore  
 foot, b—hind foot)



- b.....blood vessels  
 c.....corium  
 e.....epidermis  
 p.....stratum papillae  
 pig.....pigment  
 s. t.....prickle cell layer  
 str. c.....stratum corneum  
 str. g.....stratum granulosum  
 str. germi. stratum germinativum  
 str. l.....stratum lucidum  
 str. m.....stratum mortificatum  
 str. reticu. stratum reticulare  
 str. subcu. stratum subcutaneum



Fig. I.



*str. m.*

*str. c.*

*str. l.*

*st.*

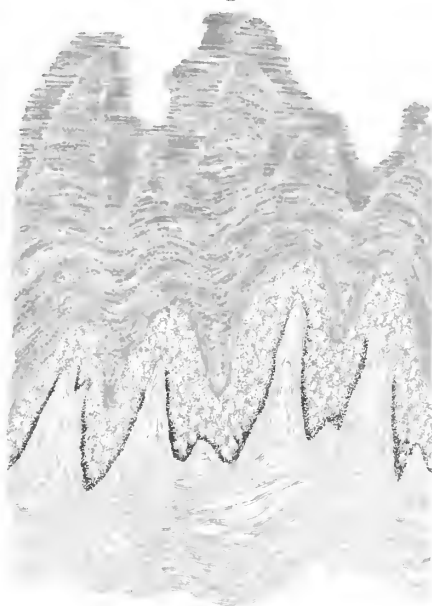
*p.*

*pig.*

*str. retic.*

*str. subc.*

Fig. II.



*str. m.*

*str. c.*

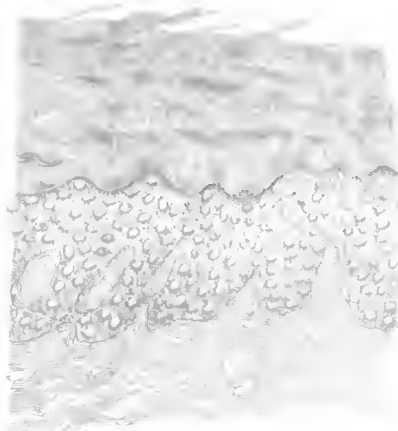
*p.*

*pig.*

*str. retic.*

*str. subc.*

Fig. III.



*str. m.*

*str. c.*

*str. l.*

*str. g.*

*st.*

*str. retic.*

*str. subc.*





Fig. I.



Fig. II.

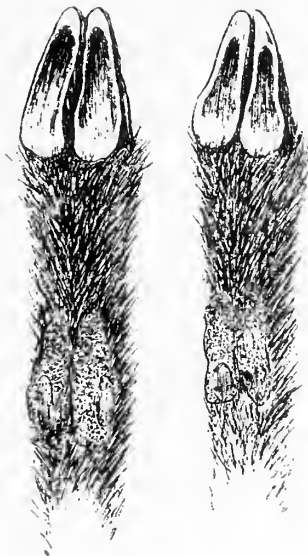
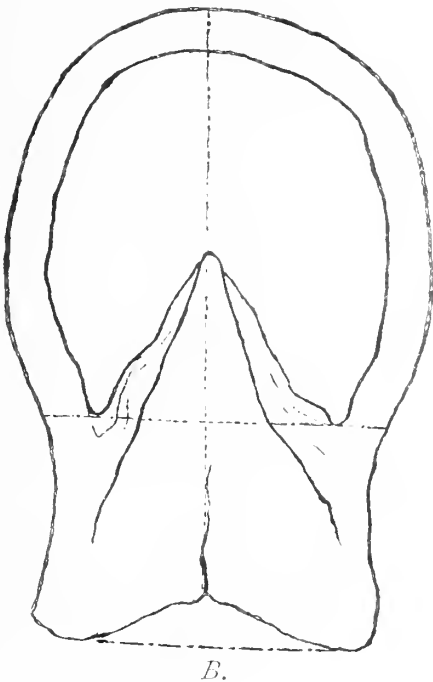
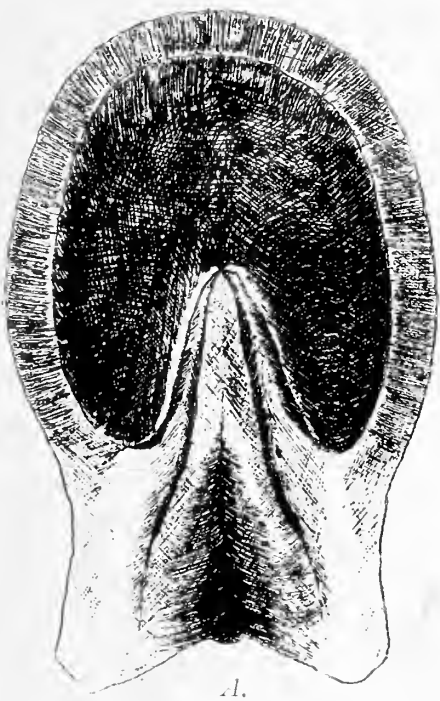


Fig. III.





# ÜBER DEN KROPF DER TAUBE WÄHREND DER BRUTZEIT

VON

**Schin. Yoschida**

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Während meines Studiums an der Universität in Sapporo veranlasste mich Herr Professor Dr. Hashimoto zu einer mikroskopisch-anatomischen Untersuchung der Verdauungswerkzeuge, insbesondere des Kropfes der Taube, um festzustellen, welchen Veränderungen derselbe während der Brutzeit unterliegt. Im zoologischen Institut zu Halle a. S. (Direktor Professor Dr. Haecker) habe ich meine Studien darüber fortgesetzt.

Bevor ich das Resultat, zu dem mich meine Untersuchungen geführt haben, niederschreibe, möchte ich Herrn Professor Dr. Haecker und Fräulein Dr. Kuttner für ihr gütiges Interesse an meiner Arbeit meinen besten Dank aussprechen. Auch Herrn Professor Dr. Hashimoto bin ich zu grossem Dank verpflichtet für das liebenswürdige Entgegenkommen, mit welchem er mir das nötige Material zur freien Benutzung überliess.

Wie schon seit langem bekannt, verändert sich der Kropf der Taube in der Brutzeit und produziert während des Auskriechens der Jungen und noch kurze Zeit darnach eine milchartige Flüssigkeit, die sogen. Taubenmilch oder Kropfmilch, die zur Ernährung der Jungen dient. Die bisher vorliegenden Untersuchungen über ihre Produktion sind in kurzem folgende: Im Jahre 1865 berichtet Hasse über seine Studien des Kropfes, insbesondere über dessen Drüsen: „Mit dem blossen Auge gesehen“, sagt er, „kann man in dem Seitenteil des Kropfes ganz feine und drüsige Falten aber keine Drüsen finden. In dem unteren Teil der Speiseröhre sind Drüsen vorhanden.“ Seine Messungen am Epithel des Kropfes ergaben, dass dieses im normalen Zustan-

de 0.1554 mm dick ist, aber bei dem Weibchen während der Brutzeit 1,5 mm, bei dem Männchen zu 2,5–3 mm Dicke anwächst. Am Schlusse seiner Arbeit weist Hasse auf den Unterschied des milchigen Saftes des Taubenkropfes und der Milch der Säugetiere hin. Diesen Unterschied untersucht Bernard noch genauer und findet dabei, dass die Milch der Säugetiere höchstens Bruchstücke von Zellen enthält, während in der Taubenmilch die abgestossenen Zellen ihre Form behalten. Nach Postma (1887) kommen Drüsen nur auf den Falten in dem Teil des Ösophagus vor, der zwischen dem Drüsenmagen und der Ausmündung des Kropfes liegt. Der Kropf ist dagegen ganz drüsenfrei.

Die speziellen Untersuchungen Teichmanns (1889) über den Kropf waren für mich von besonderem Wert, da sie mich auch mit der alten Literatur bekannt machten. Er vermutet, dass in dem Seitenteil des Kropfes Drüsen zu finden seien; doch führte die Untersuchung dieser Teile zu keinem befriedigenden Ergebnis, da er hier nur dick geschichtetes Pflasterepithel fand. In der schleimigen Flüssigkeit, die von der frischen Schleimhaut ausgeschieden wird, sah er abgestossene platte Zellen mit feinkörnigem Protoplasma und gut färbbaren Kernen. Die Färbung der Flüssigkeit liess ihn Bakterienhaufen, die auch in den Verdauungsorganen anderer Tiere zu finden sind, erkennen. Um die drüsenhaltige Stelle deutlich sehen zu können, hat er folgende Methode angewandt: „Nachdem man den Kropf und die angrenzenden Teile der Speiseröhre freigelegt hat, unterbindet man die Speiseröhre unten am Drüsenmagen und lässt von oben her Alkohol in die Speiseröhre und den Kropf einfließen, unbekümmert um die Füllung des Kropfes mit Nahrung. Wenn die Gewebe sich dadurch vollkommen ausgedehnt haben und kein Alkohol mehr aufgenommen wird, unterbindet man auch oben und bringt das Ganze in ein Gefäss mit Alkohol zu vorläufiger Härtung.“ Durch diese Methode gelangte er zur völligen Entfaltung des Kropfes und konnte feststellen, ob seine Vermutung, nach der sich Drüsen an einer engerbegrenzten Stelle befinden mussten, richtig sei. Diese Vermutung wurde jedoch nicht bestätigt. Wohl aber zeigten sich Drüsen am Übergang des Kropfes in die

Speiseröhre. Nach seiner Ansicht erstrecken sich die Leisten der Speiseröhre (6–8 an Zahl), in denen sich die Drüsen konzentrieren, 1 cm weit in den Kropf; zwischen den Falten finden sich keine Drüsen. Bei voller Ausdehnung des Organes kann man genau die Falten der Speiseröhre von denen des Kropfes unterscheiden. Die Drüsen in den Leisten bezeichnet er als zusammengesetzt schlauchförmige. Philipp Barthels kommt in seiner Arbeit über die Histologie des Ösophagus verschiedener Vögel (1895) in Betreff der Drüsen des Kropfes der Taube zu derselben Ansicht wie Hasse. Er vergleicht den normalen mit dem in der Brutzeit veränderten Kropf und findet dabei folgende Unterschiede:

Normaler Kropf bei der *Columba dom.*:

Dicke der ganzen Wand in den Falten  $816\mu$  und zwischen den Falten  $274\mu$ ; Mucosa  $147\mu$ . Inneres Bindegewebe in den Falten  $506\mu$ . Zwischen den Falten  $6.5\mu$ . Längsmuskulatur in den Falten  $98\mu$ . Zwischen den Falten  $49\mu$ . Ringmuskulatur  $68\mu$ .

Kropf der *Columba dom.* beim Füttern:

Dicke der ganzen Wand in den Falten  $4000\mu$ , ganze Schichtdicke der Mucosa  $3200\mu$ . Inneres Bindegewebe  $425\mu$  (in den Falten). Längsmuskulatur in den Falten  $245\mu$ . Ringmuskulatur  $130\mu$ . Über die Drüsen des Kropfes schreibt er, dass man auf einer kurzen Strecke welche finden könnte. O. Zietzschmann sagt in der „Vergleichenden Mikroskopischen Anatomie von Ellenberger“ Seite 1893–1894, dass die Schleimhäute des Kropfes hohe Falten aufweisen und mit dickem verhorntem Epithel bedeckt sind. Der Hauptteil des Kropfes ist bei den meisten Vögeln ganz drüsenfrei, bei der Taube gibt es keine eigentlichen Drüsen im Kropf, seine Ansicht über die Topographie der Drüsen des Kropfes deckt sich mit der Teichmanns. In der Schleimhaut der Leisten hat er auch Drüsen gefunden. Die Kropfmilch bezeichnet er, ebenso wie Teichmann, als eine weisse krümelige Masse von beissendem Geruche, der im Gegensatz zur Säugetiermilch das Kasein fehlt. Was die Produzierung der Taubenmilch anbetrifft, pflichtet er Archangelis Ansicht bei, nach der die Fettproduktion in der Nähe der Propriagefässe stattfindet.

Die Vaskularisation ist beim Weibchen entwickelter als beim Männchen, bei letzterem sind mehr colostrumähnliche Körperchen zu finden. Hasse schreibt auch über diese Frage: „Der Kropf des Männchens ist in dieser Zeit dem des Weibchens gleich, zur Zeit der Milchproduktion beträgt nämlich die Stärke des Epithels beim Weibchen 1,5 mm und die des Männchens 2,5–8 mm. Wie aus alledem hervorgeht, ist noch nicht endgültig festgelegt worden, ob der Kropf der Taube Drüsen enthält, ob die Drüsen der Speiseröhre mit der während der Brutzeit produzierten Milch des Kropfes zusammenhängen und ob während der Brutzeit der Kropf des Männchens dem des Weibchens gleich ist. Deshalb möchte ich in folgendem das Resultat meiner mikroskopischen Untersuchungen mitteilen.

#### **A. Makroskopische Anatomie des Kropfes.**

Der Kropf der Taube besteht aus zwei grossen symmetrischen Säcken an der Ventralseite des Ösophagus; die Speiseröhre des Kropfes ist beim normalen Kropf gegen die anderen Teile der Speiseröhre nach oben hin nicht deutlich abgegrenzt, nur während der Brutzeit lässt sich eine deutliche Grenze erkennen, weil dann die Falten des Kropfes eine grössere Ausdehnung haben als die des Ösophagus. Der Kropf der jungen Taube, der dem falschen Kropf der Enten und Gänse ähnelt, ist nur eine einfache Erweiterung der Speiseröhre, an der sich die Grenze noch nicht erkennen lässt. Am unteren Ende ist die Grenze gegen die Speiseröhre sowohl beim normalen Kropf, wie bei dem während der Brutzeit veränderten deutlich zu sehen, weil die Falten der unteren Speiseröhre selbst bedeutend sind. (Fig. 1) Nach meinen Untersuchungen erstrecken sich die Falten der unteren Speiseröhre (in Gegensatz zu Teichmanns Beobachtungen) nicht in den Kropf hinein; dagegen bilden die Falten der unteren Speiseröhre an der Grenze nach dem Kropf zu eine trichterförmige Erweiterung. (Fig. 1) In der Zeit während der Brut sind am Kropf die Falten, sowie die Stärke der Wand der Seitensäcke deutlich von denen der Fortsetzung der Speiseröhre zu unterscheiden; Falten und Wand der Seitensäcke sind stärker verdickt als

diejenigen der Speiseröhre und mit weisslichem milchigem Saft, der sogenannten Taubenmilch bedeckt.

### **B. Mikroskopische Anatomie des Kropfes.**

Als histologisches Untersuchungsmaterial diente mir ein Teil aus dem an die Fortsetzung der Speiseröhre grenzenden Seitensackes und ein Teil von der Grenze nach der unteren Speiseröhre hin.

Mit Zenkerscher und Flemmingscher Flüssigkeit fixierte ich dieses Material und wusch es 24 Stundenlang in fliessendem Wasser aus, darnach wurde es in 60% igem Alkohol steigend bis zu absolutem Alkohol gebracht und schliesslich in Xylol und Paraffin eingebettet. Zur Färbung benutzte ich Hansens Haematoxylin. Bei der histologischen Untersuchung zeigte sich ein Unterschied zwischen dem normalen Kropf und dem Kropf während der Brutzeit bloss im Epithel, das ausserordentlich hypertrophisch und blutreich wird, namentlich in den Seitensäcken. Diese Veränderung ist beim Seitensack bedeutender als bei der Fortsetzung der Speiseröhre. Ich habe bei dem normalen, sowie bei dem veränderten Kropf Messungen gemacht. Das verdickte Epithel der Seitensäcke beträgt während der Brutzeit 2,2 mm im Durchschnitt, bei dem normalen 0,8 mm. Die untersten Zellen des Epithels haben eine runde Form und sehr grosse deutliche Kerne, nach oben zu werden sie flacher. Die äussersten Zellen des Epithels sind bei dem normalen Kropf deutlich verhornt, aber während der Brutzeit sind hier nur viele abgestossene Zellen ohne Kerne zu finden. An dem Epithel, das sich gerade gegenüber den Papillen befindet, zeigen sich oft kernhaltige Zellen. Die Epithelzellen verfetten sich meistens, und im Epithel kann man ganz verfettete Zellengruppen finden, die mit den übrigen Zellen abgestossen werden (Fig. 4). Die unteren Epithelzellen sind während der Brutzeit noch deutlicher kernhaltig als die des normalen Kropfes, und manchmal sind hier Zellteilungen zu sehen. In der Submucosa finden in der Brutzeit keine Veränderungen statt; es sind hier aber deutlich Fasern zu bemerken. Die Schichten der Muscularis bleiben auch unverändert. Deshalb kann ich der

Ansicht Teichmanns, dass in allen Schichten Veränderungen stattfinden, nicht beipflichten. Wie Teichmann sagt, sind die Leisten des Kropfes mit den Leisten des Ösophagus nicht verbunden und deshalb leicht zu unterscheiden. Wenn der Kropf mit Alkohol oder Formalin fixiert wird, schrumpfen seine Leisten ein, dagegen bleiben die Leisten des Ösophagus unverändert (Fig. 2). In der Brutzeit werden die Leisten des Kropfes fast so gross wie die des Ösophagus. Sie sind deshalb nur nach der Richtung, die sie einnehmen, zu unterscheiden. Ihre Grenze, die eine Zickzacklinie bildet, ist immer ganz deutlich. Von den Drüsen, die Teichmann schlauchförmig nennt, habe ich sehr viele in den Leisten des Ösophagus und auch zwischen denselben gefunden. Ich widerspreche somit der Ansicht Teichmanns, nach der sich keine Drüsen zwischen den Leisten zeigen. Meiner Vermutung nach hat er einen Schnitt durch die im Zickzack gehende Grenze zwischen Kropf und Ösophagus gemacht und infolgedessen nur die Zwischenräume der Leisten im drüsenfreien Kropf gesehen. Um die Frage zu beantworten, ob während der Brutzeit der Kropf des Männchens dem des Weibchens gleich ist, habe ich viele Taubenpaare in der Brutzeit untersucht; ich konnte dabei (im Gegensatz zu Hasse) keinen Unterschied zwischen dem Epithel der beiden Geschlechter finden. Bisweilen zeigt zwar der männliche Kropf grössere Veränderungen als der weibliche, aber ebenso oft war wieder das Gegenteil festzustellen.

In der sogenannten Taubenmilch waren bei mikroskopischen Untersuchungen abgestossene kernhaltige Zellen, verfettete Epithelzellen, abgestossene Oberflächenepithelzellen und Bakterienhaufen nachzuweisen. Die letzteren sind bereits von Teichmann bemerkt worden. Auch habe ich in der Taubenmilch, die der Kropf der Jungen einige Tage nach dem Auskriechen enthält, ebensolche Zellen und Bakterienhaufen gefunden. Um zu konstatieren, ob die Taubenmilch dieselbe physiologische Wirkung hat wie das Colostrum der Säugetiere, habe ich folgendes Experiment angestellt. Ich liess Taubeneier in einem Brutapparat ausbrüten und fütterte die junge Taube bloss mit in Wasser aufgeweichten Getreidekörnern; ihr Gedeihen



dabei beweist mir, dass die Taubenmilch keine physiologische Einwirkung auf das Wachstum der jungen Taube hat. Die chemische Zusammensetzung der Taubenmilch ist eine ganz andere als die der Milch der Säugetiere; die Taubenmilch enthält keinen Milchzucker; auch ist sie, wie ich schon erwähnte, ihrer histologischen Struktur und Entstehung nach durchaus verschieden von der der Säugetiere. Hierin decken sich meine Resultate mit denen von Teichmann. Auch die physiologische Wirkung der Taubenmilch ist eine ganz andere als die der Säugetiermilch. Wenn sich im Kropf der jungen Taube diese Milch befindet, so hat das folgende Ursachen: Wie bekannt, nimmt die junge Taube mit ihrem noch weichen und sehr langen Schnabel die Nahrung aus dem Kropf der Eltern, die dieselbe darin aufgeweicht haben. In der Brutzeit kann das Elternpaar aber nicht das Nest verlassen, da es die junge Brut vor der Kälte schützen muss, deshalb enthält der Kropf nur wenig Futter, ja bisweilen ist er sogar ganz leer. Die junge Taube hat aber schon das Bedürfnis nach Speise und erhält als Nahrung den milchigen Saft.

Auch die Frage, warum sich nur das Epithel des Kropfes während der Brutzeit verdickt und nicht auch die übrigen Teile, scheint mir von besonderer Bedeutung; ich werde die Lösung dieser Aufgabe später versuchen. Wenn ich zum Schlusse dieser Untersuchungen meine Erfahrungen zusammenfasse, so ergibt sich:

- 1.) Der Kropf der Taube ist völlig drüsenfrei.
- 2.) Die Veränderung des Kropfes während der Brutzeit besteht bloss in der Verdickung des Epithels.
- 3.) Die angesammelte Taubenmilch entsteht aus den verfetteten abgestossenen und kernhaltigen Epithelzellen.
- 4.) Die Bildung der Taubenmilch geht nicht von den Drüsen der unteren Speiseröhre aus.
- 5.) Während der Brutzeit findet im Kropfe der männlichen, ebenso wie in dem des weiblichen Vogels die sogenannte Taubenmilchsekretion statt.

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**Erklärung der Abbildungen auf Tafel IX.**

- Fig. 1.) Innenseite des normalen Kropfes mit unterer Speiseröhre (Normalgrösse).  
 Fig. 2.) Innenseite des Kropfes während der Brutzeit (halbe Grösse).  
 Fig. 3.) Das Epithel des Kropfes während der Brutzeit.  
 Fig. 4.) Ganz verfettete Zellengruppe in dem Epithel des Kropfes.

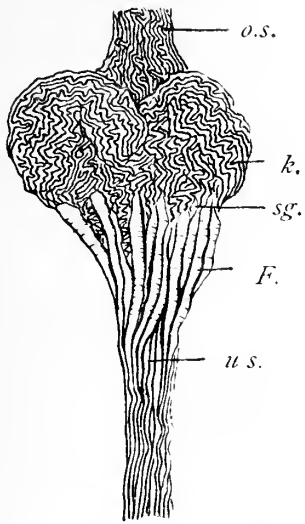
**Allgemeine Bezeichnungen.**

|                                                          |                                    |
|----------------------------------------------------------|------------------------------------|
| ab. .... abgestossene Zellen                             | b. .... Blutgefässe                |
| o. b. .... Oberflächenepithel                            | f. l. .... verfettete Zellengruppe |
| f. f. .... Falten des Kropfes                            | k. .... Kropf                      |
| o. s. .... oberhalb der Speiseröhre                      | s. b. .... Submucosa               |
| s. g. .... Grenze zwischen der Speiseröhre und dem Kropf |                                    |
| t. p. .... Tunica propria                                | u. s. .... untere Speiseröhre      |

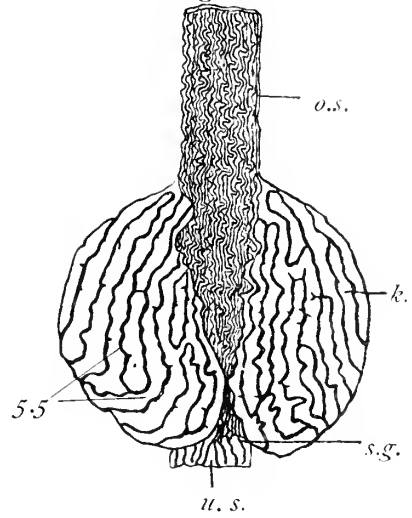




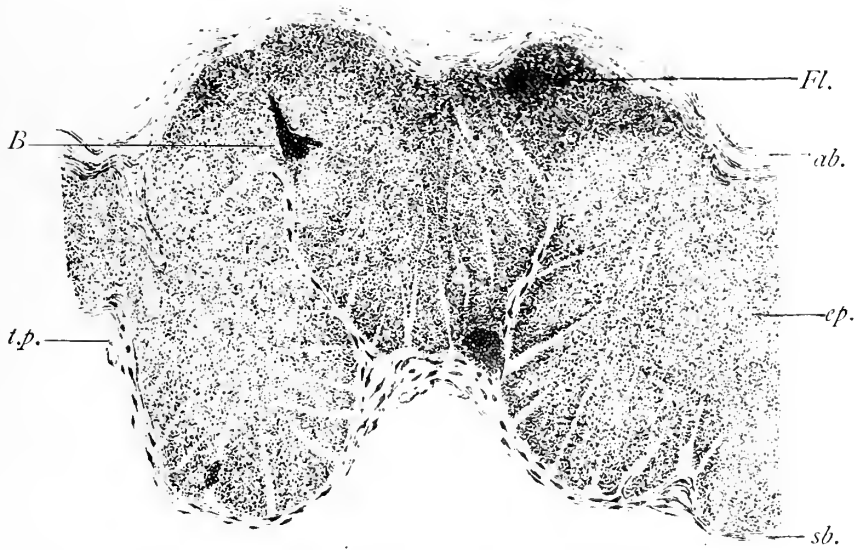
*Fig. I.*



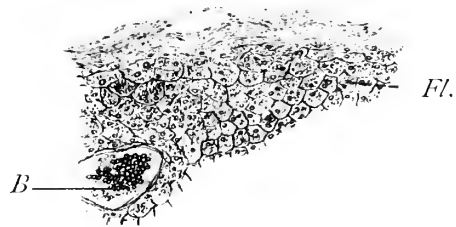
*Fig. II.*



*Fig. III.*



*Fig. IV.*





# ON THE STOMACH GLANDS OF THE RAT AND RABBIT.

By

**Schin. Yoschida.**

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In connection with the stomach glands of mammals the following points still remain undecided :

1.) The original character of the cardiac glands found in the stomach of most mammals. The results of many histologists' researches into the original character of these glands may be divided into two classes :

- a) The cardiac gland is a peculiar stomach gland like the fundus and pylorus glands.
- b) The cardiac gland is a descendant of some other gland, probably the Fundus.

2.) There are several minor questions :

- a) Are the chief cells of the fundus gland the same as those of the pylorus or are they not ?
- b) On the appearance of the Parietal cells and their histological structure.
- c) Whether different species of animals have a different distribution of the stomach glands.

This paper has been written in order to answer these questions.

In the first place I should like to give a historical review of the observations on the stomach glands.

Since, Professor Ellenberger in 1881, made his anatomical and physiological studies of the third stomach of ruminants and made clear the physiologi-

cal side of that stomach's function, the stomach gland has been studied by many anatomists.

As these studies, however, have been repeated by many histologists, I confine my remarks to a citation of the results in chronological order.

In 1836, Support Boyd was the first to study the stomach glands of the pig. He wrote that in the cavities of the mucous membrane there are no glands, but that blind sack-like glands take their place. He distinguished the Cardia and Fundus glands by their contents.

In 1838, Bischoff remarked from the histological side that there are many varieties in stomach glands, and threw some light on this subject.

In 1839, Wassmann studied the stomach of the pig, and distinguished three kinds of stomach glands: the Cardia gland, the Pylorus gland and the Fundus gland.

In 1870, Heidenhein, in the so-called Pepsin gland, distinguished two kinds of cells: the chief cells and the Parietal cells; to the glands which are in the Pylorus part he gave the name of Pylorus glands and to those in the Fundus part the name of Fundus glands.

Kölliker, found in the cardiac part of the human stomach some glands, which are distinguished by their histological structure from the Fundus glands. These glands he called the cardiac glands. He published his observations in, "Mikroskopische Anatomie, Bd. 11 (1850)."

In 1882, Ellenberger examined the stomachs of different domestic animals and wrote that the part of the stomach of many animals in which these glands are found, do not resemble to corresponding parts of the human stomach. Further, in 1885, he studied, with Hofmeister the stomach of the pig, and found besides the Pylorus and Fundus glands, still another peculiar gland in the cardiac part. This he named the cardia gland, and wrote that this cardia gland in the stomach of the pig is of a quite different histological structure from the Cardial gland which Kölliker had previously found in the human stomach. In 1887, he wrote in his handbook that he had found the cardiac gland in the narrow strip close to the Margo Plicatus.



In 1889, Edelman examined the stomach glands of various animals and, by the peculiar arrangement, direction and epithel of the gland tubes, distinguished the cardiac gland from the Pylorus and Fundus glands. He detailed many characteristics of the cardiac gland: that the cardiac gland has no Parietal cells, and that many lymph vessels are to be found in the Mucous Membrane. He further wrote that the cardiac gland is to be found in the connection of the Mucous Membrane of the Oesophagus and the digestive Mucous Membrane of the stomach, in a special bag in a separated part of the fore stomach. He found the cardiac gland in *Mus muscuraris*, *Mus domesticus* and other animals, but not in *Lepus timidus* and many other ruminants.

In 1898, Scheffer found special glands in the human stomach that correspond to the cardiac glands of animals. In this case these glands have Parietal cells, and the gland tubes are covered with cup-cells that are not affected by mucous staining, but have a colour reaction similar to that of the Pylorus and fundus glands.

In 1896, Professor Oppel published a text book, "Lehrbuch der vergleichenden mikroskopischen Anatomie der Wirbeltiere". In the first volume of this he collected all the literature on the subject up to the time of writing and formed a hypothesis for the cardiac gland as follows: "The large expansion is a sinking proceeding: the cardiac gland is not a peculiar gland". For this great work which has served as a key to all histologists who study the stomach glands, we must offer him our best thanks.

In 1902, Bensley said that the cardiac gland is a degenerated organ which has been formed from the fundus gland by the disappearance of its characteristic parts: the parietal cells and the zymogenic chief cells; also that the cells of the cardiac gland show no difference from those of the pylorus and fundus glands.

In 1907, Froehlich found in the stomach of the horse a peculiar gland, which, in its characteristics, is intermediate between the cardiac and the fundus glands. To these he gave the name of, "Übergangsdrüsen" or "Intermedial Drüsen." The histological structure of these glands resembles that of the

pylorus glands in one respect and that of the cardiac glands in many others. These glands may be stained with congo red but not with mucicarmine. The pylorus glands, on the contrary, are reacted on with Mucous stain. To distinguish these "Übergangsdrüsen" from other glands, he made the following studies: Material, not embedded in paraffin and unstained is spread out in a drop of warm water placed on the objective glass, the "Übergangsdrüsen" will then show an opaque yellow-grey colour while the Pylorus glands show a dark grey-blue shade.

He further studied the stomachs of goats, cattle and pigs, and found many varieties of "Übergangsdrüsen" in these. In the Mucous Membrane of the dog's stomach which forms the transition to the Oesophagus, he found three varieties of glands: the Seroese and Mucoese cardiac glands and the true Fundus glands. These glands generally have Parietal cells. The Seroese and Mucoese cardiac glands can be stained with Mucous stain to the blind end, the end part of the gland is very little coloured, the reaction on a narrow side part being very good.

In conclusion he said that the histological structure and chemical contents are not the same, but that these glands are to be considered as a phylogenetical simplification and are much simplified descendants of the Fundus glands.

In 1905, Hamme wrote of the cardiac glands of the stomach of domestic mammals and distinguished them from other stomach and oesophageal glands as follows: The pylorus gland is generally straight, and broadened at the blind end while the cardiac gland has a snake-like form, no broadened end and runs singly. He also made a comparison between the cells of the cardiac, pylorus and fundus glands, but as I wish to compare his results with my own histological studies I will write of these later.

Besides these he made chemical studies and said that the fermentative contents of the cardiac gland are amylolytic, but the rennet, milk-acid, fat and peptic ferment are absent. In conclusion he wrote that the cardiac gland is by its chemical and histological structure quite different from the pylorus and

fundus glands, and that, therefore, the cardiac gland is quite distinct from all other stomach glands.

As I have said above, the opinions on the cardiac gland, up to the present time, may be divided into two classes :

1. The cardiac gland is a peculiar kind of stomach gland. This opinion is held by Ellenberger and other histologists who studied in his Institute, as Hamme and Edelmann.
2. The cardiac gland is an outcome of the Fundus gland. Professors Oppel, Fleischmann and Bensley hold this opinion.

For the sake of convenience, *Mus muscuraris* and *Lepus cuniculus* have been employed as material. From these animals which have been killed with chloroform vapour I have cut out the stomach with a small portion of the lower part of the Oesophagus, a small portion of the upper part of the small intestine remaining. It was cut open along its long curvature and the contents washed away with physiological salt solution. The stomach was divided as follows :

1. A connected part containing the thick-walled portion which will be called the cardiac sac and the thin-walled portion, the Fundus portion.
2. A connected part containing a portion of the small intestine and stomach proper and other different portions.

This was thrown into fixation fluids. As fixation fluids I have always used Picro-sulphuric acid solution, Zenker's solution and Muller's fluids.

The material was placed in 70% alcohol, then transferred to 80%, and to successively higher grades of alcohol. As staining I have used double staining, Hansen's haematoxylin combined with congo-red. For Mucous reaction of the stomach gland, I have used muci-carmin, iron-alum, eosin and toluidin. The material was cut in serial sections with a thickness of 5 $\mu$ .

In 1909, I studied the stomach of the *Mus muscuraris* and *Lepus* from a histological and anatomical standpoint, but since that time I have obtained further material and continued my researches, the results of which are given in this paper. Whenever my present results correspond with my former

ones, I shall repeat these.

### A. Macroscopical Features.

The stomach of the rat is reniform in outline and receives the distal end of the Oesophagus at the middle point of its concave surface ; the half to the left of the oesophageal connection is raised and bent against the oesophageal tube, while the right half passes over through the pyloric constriction into the Duodenum. The left half is thick-walled and shows in fresh specimens dully white colour, resembling in these respects the Oesophagus ; the right half has, on the other hand, comparatively thin walls which are, like the Duodenum, of pink colour.

On the inner surface of the stomach likewise two portions are to be distinguished ; the thick-walled portion which will be called the cardiac sac simply because it represents the cardiac portion in other mammals, and the thin-walled portion comprising these parts which represent respectively the Fundus and the pylorus. This latter portion will be called the Ventriculum. The surface of the cardiac sac is very harsh and leather-like, owing chiefly to the presence of a complicated system of wrinkles, causing the walls to be thickened. The wrinkles which, though variable, occur mostly from seven to eight in number are undulated in their course ; nevertheless, each two, or sometimes three of them run in parallel. The wrinkles in the proximal part pass over into the corresponding structure of the Oesophagus.

The wrinkles take in the Oesophagus not an undulated course as in the cardiac sac, but are represented by a number of longitudinal ridges which are nearly uniform in thickness and cause the oesophageal walls to be thickened. Whilst none of these longitudinal ridges are uninterruptedly continuous with the cardiac wrinkles, others disappear at the neck by which the Oesophagus is connected with the stomach.

Killing the animal immediately after feeding and cutting its gastric pouch open ; we find the cardiac sac filled up with the food roughly masticated, while the Ventriculum remains empty. The cardiac sac is therefore

to be looked upon physiologically as the food-store, similar to the rumen in Ruminants.

From the statements above given it is true that the cardiac sac can not be distinguished from the Oesophagus by means of a rough macroscopical survey such as just done, so that the two parts are to be regarded as one and the same structure. In other words, the cardiac sac is nothing other than the distal part of the Oesophagus strongly bulging out.

The Fundus, the broadest part of the stomach, is marked off against the cardiac sac just stated by a continuous ridge describing a circle which is situated in an oblique position. On the bottom surface of the stomach the boundary ridge is found just opposite the oesophageal opening, and on the roof surface, where the circle is notched, it is seen right to, and at a little distance from the oesophageal opening, while on the lateral walls it is shifted to the left side beyond the oesophageal opening. The Oesophagus is, therefore, not led directly to the Fundus, but passes into the cardiac sac through a narrow groove which deserves the name of Oesophagus-cardiac groove. The boundary ridge itself shows numerous large and small undulations which occur especially in the adjacent parts of the oesophageal opening. The surface of the Fundus walls is apparently smooth; in fixed specimens some irregular foldings appear, owing doubtless to a strong contraction of the muscle-coat of the stomach.

Without any demarcation, the fundus passes over into the Pylorus where the gastric sac is considerably narrowed into the pyloric constriction. The surface of the pylorus walls is likewise smooth. The Duodenum which is represented by a slender tube forming the distal continuation of the Pylorus shows a sudden change on the inner surface, presenting velvety appearance, which is, of course, due to occurrence of the thickly set together

In the stomach of *Lepus cuniculus* (rabbit) the cardiac and pylorus portions may be distinguished on the surface of the inner side, the surface of the latter portion being tufted; the wall on the former is rather thin, particularly when the stomach is full of food. Food, when taken, passes into the cardiac

portion and is kept here, then gradually sent into the pylorus and fundus portions as we have already seen in the case of the stomach of the rat.

The cardiac portion of *Lepus cuniculus* is very large, occupying two-thirds of the stomach. The connection of the cardiac and fundus glands is not distinguishable by the naked eye and there is no boundary ridge.

### **B. Microscopical Anatomy.**

According to the above mentioned modification of the gastric pouch, the three layers of which the gastric walls are composed show features peculiar to each of the three regions. Most striking are the changes undergone by the innermost layer of the three, the Mucosa layer. Fig. 1. represents a semidiagrammatic longitudinal section through the boundary ridge which marks off the Fundus against the cardiac sac. The regular Epithelium which lines the inner surface of the Fundus passes over, at the point marked with X, suddenly into a multicellular Epithelium, lining not only the boundary ridge itself, but the cardiac sac also.

The Mucous Epithelium is uninterruptedly continuous with the multicellular Epithelium; yet these two sections of the Epithelium are widely divergent from each other as regards their histological character. The former is constructed of a single row of columnar cells taking regular epithelial arrangement; the latter is, on the contrary, composed of a layer many cells deep and accordingly attains a considerable thickness in contrast to the comparatively thin Mucous Epithelium. In this multicellular Epithelium, moreover, two layers are distinguished; the outer layer is, on sections, fibrous in appearance, while in reality it is formed of numerous strata of dry flattened epithelial cells loosened to be ultimately cast off; the inner layer is compact in texture, being composed of protoplasmic epithelial cells closely set together. In this compact part of the Epithelium several interesting facts are to be noticed. As will be seen on reactions, the nuclei which are scattered in the layer in question are thickly put together in the basal part of it and grow lesser and lesser towards the peripheral part where the layer shows every

appearance to be split so as to give rise to the outer loosened layer. In the basal part the nuclei are crowded because here active cell-multiplication is taking place.

From the facts above pointed out, it is true that the cardiac sac which occupies in volume about one half of the gastric pouch cannot be distinguished histologically from the Oesophagus, it represents a part of the Oesophagus. We see, therefore, that the results arrived at by the microscopical examination perfectly confirm my conclusion above given according to which the cardiac sac is nothing but a part of the Oesophagus strongly bulging out and serving as a temporary strage place for food. If this assumption is valid, the boundary ridge itse'f is to be included in the esophagal parts ; for it is by no means distinguishable from the cardiac sac as far as the microscopical structure is concerned.

Let us now turn to the corresponding structures in the domestic rabbit, which have been almost satisfactorily made known by previous authors. In the first place, the Gastric pouch is simple, being entirely occupied by the ventricular part ; there is no appendage existing which corresponds to the cardiac sac of the rat. As obvious, the left part of the gastric pouch is protruding upwards, apparently looking like a cardiac sac, but it is a part of the ventricular section so raised. Consequently the boundary ridge shifts to the so-called Cardia at the base of the Oesophagus, describing a small circle at the height of the Gastric pouch, instead of a large ellipse at the middle transverse vertical plane of the corresponding pouch in the rat. As a comparison of (Pl. X. Fig. 2) which represents a diagramatic longitudinal section with (Pl. X. Fig. 3) which is a diagramatic view of the median longitudinal section through the rabbit's stomach and the Oesophagus shows, the difference is striking between the gastric pouches of the animals referred to ; if the stomachs of these two animals are taken as being equal in bulk, the digestive surface of the rat's stomach is in extent only half of that of the rabbit.

As the necessary consequence of such a gastric construction as above given, the distribution of the gastric glands of the rabbit shows correspond-

ing divergence in the rat's case.

### (I) The Cardiac Glands.

The cardiac glands of the stomach of the Rat are distributed in the narrow zone just inside the boundary ridge so as to describe an ellipse along the latter. The cardiac glands of the rat, according to the many specimens I have examined, are in contradiction to the assertions of many histologists never more than seven in number. I, at least, have never been able to find so many. The connection between the Epithel of the cardia gland and the Epithel of the Oesophagus that composes the cardiac sac, is the communication between the epithelium cells of the cardiac gland and the cells which have a relatively large nucleus that shows a good reaction with Haematoxylin and which envelope the Papillae of the mukose, these having a complex form in the connecting part. The change from the epithelium cells of the cardiac gland and the basal cells of the Epithelium of the cardiac sac in the region of the junction is very gradual and the two cannot be distinguished.

The form of the cardiac gland is not branched and shows a very simple shape. The tubes run first at right angles to the wall of the stomach and then parallel or slightly inclined, and parallel to the ridge which forms the connection with the cardiac sac.

The gland nearest the connection with the Oesophagus is very simple, sometimes formed of a single cavity. The epithelium cells of the cardiac gland are of homologous columnar form and are firmly joined together. The nucleus lies at the bottom of the cell. The surface epithelium cells of the cardiac gland are of higher cylindrical form than those at the bottom, but the contents of all cells of the gland are the same.

The connection of the cardia and fundus glands may be known from the appearance of parietal cells. The cells of the cardiac gland gradually change to the chief cells of the fundus gland; in this place, therefore, the cells of some of the glands show an intermediary form between those of the cardiac gland and the chief cells of the fundus gland. Among such intermediary



cells parietal cells may sometimes be found.

In the case of the pig and the horse, other histologists have obtained no colour reaction at all, but in the case of the rat a weak mucous reaction is the result which, with eosin, is still better.

The cardiac glands of the stomach of hares and rabbits are distributed over a small area, being arranged in a row at the base and along the boundary ridge; they are consequently much fewer in number as compared with those of the rat, but they can be distinguished from the fundus glands by their histological structure. The cells of the cardiac glands are quite similar to those of the cardiac glands of rats.

As written above the number of cardiac glands is very small, sometimes, therefore, intermediary glands may be seen between the Oesophagus and fundus glands. From my own observation, I have never found more than four rows of cardiac glands along the connection of the Oesophagus and the stomach.

Hinze distinguished the cardiac gland cells, the chief cells of the fundus gland and the pylorus gland cells by their histological structure and wrote, "The cells of the Pylorus gland are flat and have a nucleus at the base and show a good reaction with mucous colours (muci-carmin and toluidin-blue,) similar to the cells of the mucous gland. The cells of the cardiac gland have relatively larger and blister-like nuclei; no reaction is obtained with mucous colour.

The difference between the fundus and the cardiac glands is that parietal cells are found in the former; the cardiac glands are affected by eosin, the chief cells of the fundus glands, however, very little or not at all."

I shall have occasion, later on to write my own views on the histological difference between the cells of these three glands.

## (II) The Fundus Gland.

The Fundus portion of the rat is next in size to the cardia sac and is connected with this by the cardiac glands. The tube of the fundus gland may

be divided into two parts according to form and histological structure, viz: the gastric crypt and the gland lumen. The epithelium cells of the Gastric crypt are, on the superficial epithelium, of elongated cylindrical form and firmly joined together, but they gradually become rounder and the contents increase towards the end part of the gland. The cells which enclose the end part of the gland may be distinguished from those of the gastric crypt as the former are of very round form with the nucleus in the middle, while the contents are much clearer than those of the latter. When stained with hæmatoxylin the cells of the epithelium and of the gastric gland are more coloured than those of the end part of the gland. When eosin is used for staining the cells of the epithelium show some reaction, the chief cells of the cardiac gland give a very good reaction. With mucous colour some reaction is obtained in the cells of the epithelium and in the chief cells of the fundus it is very good, while in the cells of the cardiac gland there is occasionally a very slight reaction but usually none at all.

The result of the observations of colour reaction in the cells of the epithelium and in the chief cells of the fundus gland may be summarised as follows: with mucous colouring the cells of the cardiac gland and those of the fundus gland are affected in a quite opposite manner, and the cells of the epithelium of the fundus gland have a similar colour reaction to those of the cardiac gland.

Among the intermediate cells between the gastric Crypt cells and the cells of the end part of the gland I have found Parietal cells. These, as Heidenhein and other histologists have remarked, are of round or oval form with the nucleus in the middle, and the ends of those against the Lumen pointed. My finding Parietal cells among the intermediate cells agrees with the result of Heidenhein's observations (1870), when he found Parietal cells among the cylindrical cells of the gland outlet of the Fundus. Bentkowsky (1796) found Parietal cells in the outlet of the Pepsin gland and Oppel obtained a similar result in the case of a number of mammals. In the rat, however, the Parietal cells among the intermediate cells are very few and difficult to find, and

among the epithelium of the Fundus gland I have never seen any.

In the delicate structure of the Parietal cells I have seen no network as other histologists affirm, and the contents of the cells are always homologous.

Parietal cells with more than one nucleus, which Trinkler found in 1884, and in particular the intermediary cells between the chief cells and the Parietal cells, which Edinger found in human beings in 1879, I have not been able to find.

I have seen some Parietal cells which were slightly changed by the loss of some of their contents and therefore resemble the chief cells of the Fundus, but, by staining, these can be distinguished from the chief cells. In the neck of the Fundus I have not been able to find chief cells, being enclosed with intermediary cells as related above.

By histological observation I have distinguished the different points of the chief cells of the Fundus gland and the cells of the Epithelium as follows:

The cells of the Epithelium are of high cylindrical form and firmly joined together. The nucleus always lies at the base and the cell membrane is very distinct. With hæmatoxin and eosin the cells of the Epithelium are more affected than the chief cells of the Fundus. The form and colour reaction of the cells of the Epithelium are similar to the cells of the Cardia gland. On the other hand, the chief cells of the Fundus gland have a more or less spherical form, with clear granulated cell contents. There is little or no reaction with eosin and hæmatoxin. They enclose the end part of the Fundus gland and contain many Parietal cells among them.

The Parietal cells with two nuclei, which, in 1895, Bohm and Davidoff found in this animal, I have never been able to find. I have seen, however, cases which, when stained with a strong congo-red, have a comparatively thin membrane; the contents give a good reaction so that the wall between the two cells is sometimes invisible and the two appear as one cell containing two nuclei. When a weak staining is used with such cells, the membrane between the two can be distinctly seen.

The results of the above histological observations may be summarised as

follows :

1.

The cells of the Fundus gland may be divided into four kinds :

- a. Superficial epithelium cells.
- b. Intermediate cells.
- c. Chief cells.
- d. Parietal cells.

2.

Among the superficial epithel cells there are no parietal cells.

3.

parietal cells with two nuclei are not to be found.

4.

Among the intermediate cells the parietal cells are rarely found.

5.

Intermediary cells between the chief cells and parietal cells are absent.

6.

The histological structure of the superficial epithelium cells of the Fundus gland is similar to those of the cardia gland.

The Fundus glands of hares and rabbits occupy two-thirds of the stomach surface. The cells of the fundus glands, in this case, are divided into four kinds : superficial epithelium cells, intermediate cells, chief cells and parietal cells.

The chief cells are not affected with eosin or only very slightly, but the superficial epithel cells give a very good reaction ; the intermediate cells give a correspondingly medium reaction. In this case the parietal cells are very large and occur in greater numbers among the intermediate cells than in the case of the rat. Among the superficial epithel cells, as in the rat, none are found.

In 1879, Langley and Sewell divided the surface of the stomach into four regions :

- 1.) Fundus (the gland showing comparatively few parietal cells).
- 2.) Large curvature.
- 3.) Small curvature.
- 4.) Pylorus.

I, however, have not been able to find such regions by the histological

structure, particularly is there no difference between the first and second regions.

### (III) The Pylorus Gland of the Rat.

Then next come the Pylorus glands, as they are commonly called, under consideration. The walls of the glands are composed of tall cylindrical cells, arranged regularly and compactly put together, just like those in the cardia gland. In the majority of mammals the glands send off branches at their basal part, and those of the second order which are more or less contorted pass over without sharp demarcation into the duodenal gland. In the species in question, the pyloric glands are, on the contrary, straight, and show not even a tendency of ramification, consequently the glands push, into the submucosa layer, but are confined to the outside of the muscularis mucosa stratum. In this respect the pyloric glands are very easy to distinguish from the cardia glands which bear a great resemblance to them and show sharp contrast to the duodenal glands with which they are in close annex.

The cells of the pylorus gland may be divided into two kinds: the superficial epithelium cells of the pylorus gland, which are tall cylindrical cells compactly bound together, and the cells which enclose the branched end part which are somewhat apherical and bear clear contents.

The colour reaction of the former cells is similar to the superficial epithelium cells of the fundus glands, and that of the latter to the chief cells of the fundus gland.

There is no difference between the histological structure of the superficial cells of the fundus and pylorus glands.

In 1894, Sappey obtained a similar result.

In 1869, Klein found Parietal cells among some Pylorus glands I, however, have not been able to find such in this case, but in the intermediary form glands between the Pylorus and Fundus which are found in connection with these two glands, I have sometimes seen them.

In 1870, Ebstein, gave to this same gland in the dog and cat, the name

of Magen schleimdruesen, and wrote: "The cells of the stomach surface and the epithel cells of the gland tube and the chief cells of the pylorus glands are of different structure and the latter are enclosed with chief cells of the rennet gland."

Similar to this result, in 1876, Bentkowsky wrote that the chief cells of the fundus and pylorus glands are a morphological homology.

On the other hand, Langley and Sewell (1879) stated that in the dog and the rat the pylorus gland cells are, in a fresh condition, very clear and have fine granulated contents with a generally distinct Lumen. Therefore, in a fresh condition, they may generally be distinguished from the Fundus gland cells.

In 1880, Heidenhein stated that, in a fresh condition, the chief cells of the fundus glands contain deep dark nucleuslike contents, but the cells of the Pylorus gland have much finer granulated contents.

Ellenberger, in 1884, wrote that the cells of the pylorus gland are similar to the chief cells of the fundus gland, but are not the same. He also distinguished the cells of the fundus gland from the superficial epithel cells by the fact that the former are totally coloured with carmin but that only the nucleus of the latter is affected. He also distinguished the chief cells of the fundus gland from those of the pylorus gland in that the pylorus gland cells are closely and finely granulated packed, compactly and that they contain some mucin and are easily affected by acetic acid.

According to my own histological observations, the form and the contents of the cells at the end part of the pylorus gland and the chief cells of the Fundus gland are similar, but a still more detailed comparison shows that the cells of the former are of more columnar shape and are more uniformly arranged than the chief cells of the fundus gland.

The pylorus glands of rabbits occupy one-fifth of the gastric pouch. In regard to the areal proportion of their distribution this class of glands is therefore not markedly different from that in the rat.

#### (IV) The Duodenal Glands of the Rat.

The histological structure of the Duodenal glands is very simple. They cannot be distinguished from the pylorus glands. The tall cylindrical cells of which the gland walls are composed are uniform and take regular epithelial arrangement, supported upon the basement membrane. The Duodenal glands open between the villi, at the basal part of them. Abundant occurrence of the Goblet cells in the mucous epithelium of the Duodenal marks off this section of the alimentary canal from the Pylorus, where no trace of this class of the epithelial components is met with.

The duodenal glands are lost from sight at the opening of the bile-duct.

As to the structure of the Duodenal glands of rats there are two points which perhaps need special mentioning ; the Duodenal glands in the rat show in the first place every graduation of the morphological transition to the pyloric glands, so that the Duodenum and pylorus present no sharp demarcation in this respect, whereas we see, in the rat, a sudden change in the passing from one of these parts to the other. This striking feature in the alimentary canal of the rat is, in the second place, attributable chiefly to repeated ramification of those Duodenal glands lying close to the Pylorus ; on the other hand, in the rabbit this suddenly increased ramification of the glands is not the case, as the glands are almost equally ramified throughout the whole extent of their occurrence, being neither suddenly decreased in their number, though this is the case in the rat's Duodenum.

### **Histological Review of the Researches on the Stomach of Rat and Rabbit, and Conclusion of the Macro. and Microscopical Studies.**

In spite of the rapid progress which morphological science has recently undergone a detailed study of the rat's alimentary tractus has been neglected for more than twenty years ; so far as I am aware, since the appearance of the notable observation by Edelmann in 1889, no work concerning the system of the organ in question has been published.

The first investigation was made as early as 1807, when Home published his work. This work must, of course, be appreciated so far as it is a work of such an early date ; yet it is very rough and incomplete in observation. The author incorrectly assumes that two portions, the Ventriculum and the Cardia, are found in the stomach of the house rat. In 1854, Leydig published his work which shows to a great extent full agreement with the present results obtained by myself. He correctly made out the distinctive difference of the cardiac portion from the Ventriculum. According to him, the latter portion is thickly covered with innumerable gastric glands, whereas the cardiac portion is entirely destitute of these glands, being completely covered by a consistent skin quite like the membrane coating the inner surface of the Oesophagus.

In 1889, Grimm wrote that the blind sac of the Cardia part of the mouse's stomach has plaster epithel and is lacking in glands.

The accounts given by Edelmann (1889) are, in accuracy as well as in correctness doubtless lifted above the level arrived at by all the previous observers. Whilst in the works previous to his, only the common name of the animal employed as material was given, Edelmann states the scientific name of the animal forming his material ; he made use of *Mus domesticus*. Furthermore the author pointed out, for the first time, the existence of the cardiac glands in the mouse, which, before his time had been entirely overlooked. In details, however, I cannot confirm his results. In the first place, as seen in a figure given by the author, the distribution of the cardiac glands is exceedingly large in extent ; it is so large that the glands extended so much



further as to be put in rows continuous not only with the fundus glands but also with the pylorus glands. In reality, as stated in the foregoing pages and as seen in the diagrammatic view given in (Pl. X. Fig. 2), the cardiac glands are confined to a narrow belt running along the boundary ridge marking off the Ventriculum from the cardiac portion. Edelmänn believes, in the second place, that the cardiac glands, the fundus glands as well as the Pyloric, are, as regards their distribution, different according to the species of the rats worked with by him, i. e. *Mus documanus* and *Mus rattus*. This is in reality not the case; my present work has been carried on chiefly with *M. documanus*, and to control the facts obtained, I made use of *M. rattus* but was unable to detect so remarkable a difference according to the species as Edelmänn says, though repeated careful observations were made. In the third place and lastly, there is a wide gap between the results obtained by myself and those by Edelmänn, concerning the transition of the pyloric portion to the Duodenal portion. Edelmänn gives no accurate account of the structure of the Pyloro-duodenal passage. As given in the foregoing pages, the Duodenum of *M. documanus* shows a striking difference in the pylorus on account of enormous aggregation of the Duodenal glands at the commencement of the intestine, while in *M. rattus* the pylorus gradually passes over into the Duodenum without any sudden change in structure.

In 1890, Schlavunos writing on the plaster epithel of the cardiac portion of the mouse's stomach said that the cells lying deepest are more or less round and become flatter and flatter towards the top; the nuclei show from two to six grains.

It was in 1891 that a joint work by Klein and Verson appeared. The authors believe that the Oesophagus leads directly into the cardiac portion; they consider the latter portion as a part of the former. Otherwise the work adds no facts to the results arrived at by previous authors.

The stomach glands of rabbits have already been studied by many histologists. In this place, I will note the chief observations only.

In 1807, Home was the first to study the stomach of *Lepus timidus* and

*Lepus caniculus*, and to distinguish the cardiac and pylorus portions. He stated that the Mucous Membrane of the former is thick and the surface tufted.

In 1871, Rollett studied the stomach from a physiological side, and wrote that it is impossible to find a perfectly empty stomach among rabbits, and that even when fed with milk only, solids are to be found in the stomach.

Langley and Sewell, in 1879, divided the Stomach of *Lepus caniculus* into four regions.

In 1889, Edelmann found that in *Lepus timidus* the cardiac glands are absent. But as written above, I have, in the junction of the Oesophagus and the fundus parts, found a few cardiac glands. He further says on the Fundus glands of hares, "The fundus glands have large cells which in many cases are triangular or rounded on one side and contain large nuclei. They are not affected by Eosin staining and only show a weak red colour." He could find no cardiac glands in *Lepus caniculus*.

Because specimens are so easy to obtain, many results have been gained by histologists which generally point to the same conclusion and I will therefore not quote them further.

In the foregoing lines, the chief points of structural difference of the gastric pouch of the rat from the rabbit's has, as I believe, been pointed out, and we will now turn to the summing up of

**I) The characteristics of the rat's Stomach and the Duodenum, as follows:**

- 1.) The stomach is divided into two parts equal in capacity: the proximal part constitutes the cardiac sac and the Distal part answers to the Ventricle in other mammals.
- 2.) The cardiac sac is distinguishable from the Oesophagus as regards its microscopical as well as macroscopical structure; it is represented by a basal expansion of the Oesophagus.
- 3.) The boundary ridge which marks off the two divisions of the sto-

mach from each other is oesophagal in its microscopical texture and is to be looked upon as a part of the cardiac sac.

- 4.) The distal or ventricular part of the stomach is, as in other mammals, to be subdivided into two parts by the nature of the glands occurring in respective parts. The proximal part represents the fundus wherein open chiefly the Fundus glands; the distal part constitutes the Pylorus possessing the so-called Brunner's glands. The latter part is very small in extent as compared with the former.
- 5.) The three kinds of glands discernible in the rat's stomach are distributed, but not intermingling over three zones well marked off against each other. The cardiac glands occur along the boundary ridge in a narrow zone, about three glands in breadth, describing an ellipse. Whilst the fundus glands do not differ from those in other mammals, the Pylorus glands show a remarkable contrast; they are neither branched nor crooked, nor do they enter the Submucosa layer, being represented by simple slender tubular glands.
- 6.) In the two species, *Mus muscuraris* and *Mus documanus*, I have been unable to find a different arrangement.
- 7.) The superficial epithelium cells of the Fundus gland and the superficial epithelium cells of the Pylorus have the same histological structure and are very similar to the cardiac gland cells.
- 8.) The duodenal glands show a great contrast to the pyloric glands and thereby the two parts of the alimentary tractus are sharply separated from each other; the duodenal glands are repeatedly branched, not only so but much crooked, so that the Submucosa layer containing them is considerably extended in thickness. This feature is, however, to be seen only in the beginning of the Duodenum; the canal shows a sudden decrease in branching and consequently in thickness in its submucosa layer.

**II) The characteristics of the rabbit's stomach are as follows :**

- 1.) The surface of the rabbit's stomach may be divided into two parts : the fundus gland region which occupies two-thirds of the whole and the pylorus gland region which occupies the remainder.
- 2.) The stomach glands of the rabbit like those of the rat may be divided into three kinds : the cardiac glands which can always be found in a small number in the junction of the oesophagus and stomach, the Fundus glands and the pylorus glands.
- 3.) The stomach glands of rat and rabbit have the same histological structure, only the number of the glands and the size of the cells is different.
- 4.) The arrangement of the stomach glands and their histological structure are the same.

As written above, I have, in the rat and rabbit, found three kinds of glands in the stomach, but from the histological structure of the cells of each gland five kinds may be distinguished :

- 1.) Parietal cells.
- 2.) Chief cells of fundus gland.
- 3.) Chief cells of pylorus gland.
- 4.) Superficial epithel cells of fundus and pylorus glands.
- 5.) Cardiac gland cells.

In these five different kinds of cells, the second and third are very similar, but in the rabbit's stomach I have found the slight difference, that the chief cells of the pylorus gland are more columnar and compact than those of the fundus gland, that the nucleus in the former always lies in the basal part of the cell but in the latter it lies comparatively in the middle part of the cell.

Between the fourth and the fifth kind the histological difference is very slight, particularly when the cardiac glands are very few as in the rabbit, they are then almost the same as the superficial epithelium cells of the stomach.

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**Explanation of Plates.****Plate X.**

- Fig. 1. A section from a series of sections cut vertically through the boundary ridge and its adjacent part of the rat's stomach; magnification Zeiss d. 3 and camera.
- Fig. 2. A diagrammatic view of the rat's stomach reconstructed from series of the sections. The discontinuous transverse lines show the extent of the structures of oesophageal nature; the boundary ridge is represented by the oblique lines; the crosses indicate the area in which the pyloric glands are distributed; and the area of the distribution of the fundus glands is shown by the dots.
- Fig. 3. A diagrammatic view of the rabbit's stomach.
- Fig. 4. Cardiac gland of rat.
- Fig. 5. Fundus gland of rat.
- Fig. 6. Pylorus gland of rat.

**Plate XI.**

- Fig. 1. A section from the same series of sections shown in Plate X. Fig. 1. being highly magnified; magnification Zeis D 3 and camera.
- Fig. 2. Section through cardiac gland of the rat's stomach.
- Fig. 3. Section through fundus gland of the rat.
- Fig. 4. Section of rabbit's oesophagus, showing a cardiac gland and fundus gland.



The following abbreviations are used throughout the plates.

|            |                    |
|------------|--------------------|
| a. c. .... | accessory cells.   |
| b. r. .... | boundary ridge.    |
| c. c. .... | chief cells.       |
| c. g. .... | cardiac gland.     |
| c. s. .... | cardiac sac.       |
| d. ....    | duodenum.          |
| d. g. .... | duodenal gland.    |
| e. ....    | epithelium.        |
| f. ....    | fundus.            |
| f. g. .... | fundus gland.      |
| g. f. .... | gastric.           |
| l. u. .... | lumen.             |
| m. m. .... | muscularis mucosa. |
| o. e. .... | oesophagus.        |
| p. ....    | pylorus.           |
| p. c. .... | parietal cells.    |
| t. p. .... | tunica propria.    |
| v. ....    | vilus.             |





Fig. I.

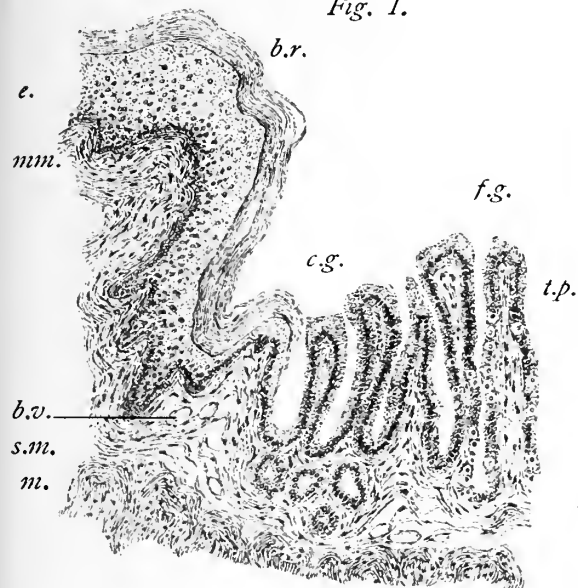


Fig. II.

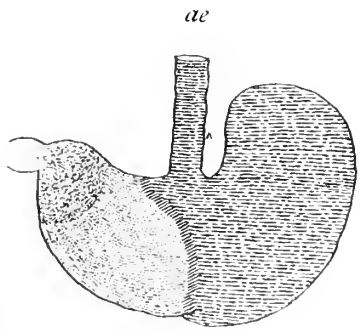
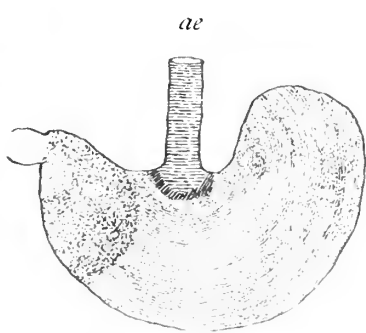


Fig. III.



- oesophagus and cardiac sac.*
- cardia gl.*
- fundus gl.*
- pyloric gl.*

Fig. IV.

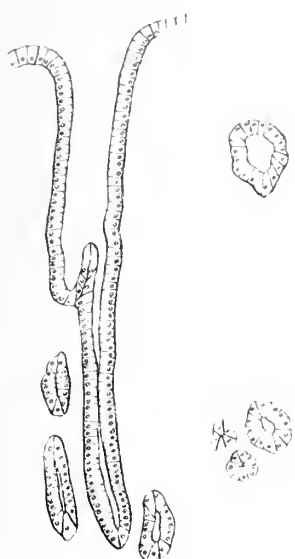


Fig. V.

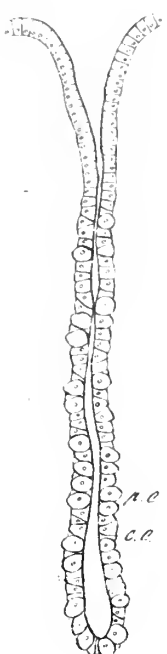


Fig. VI.

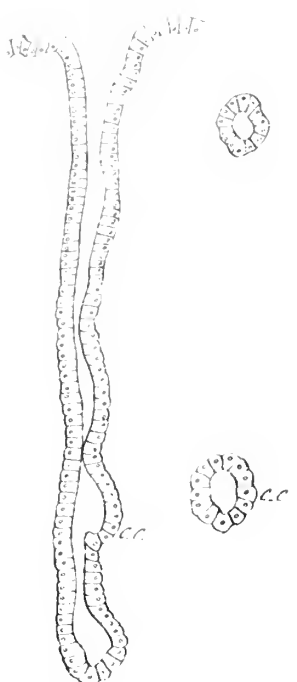




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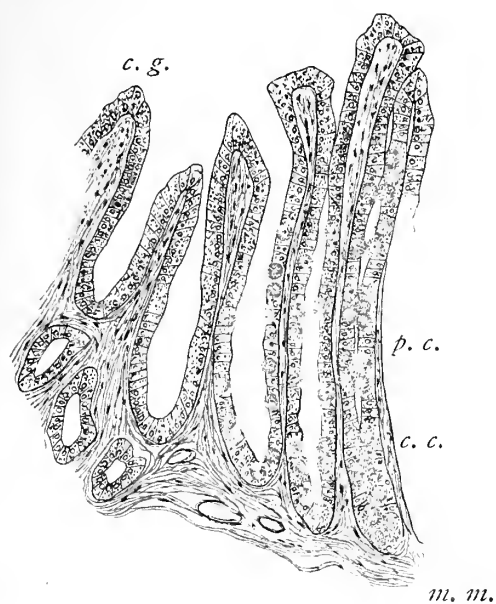


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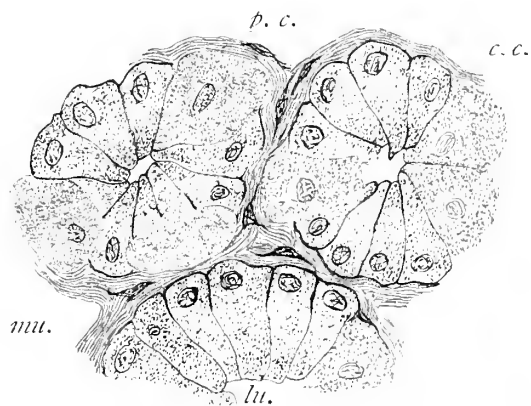
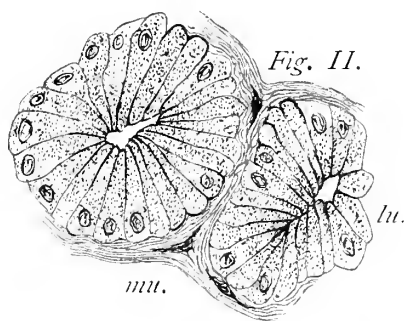
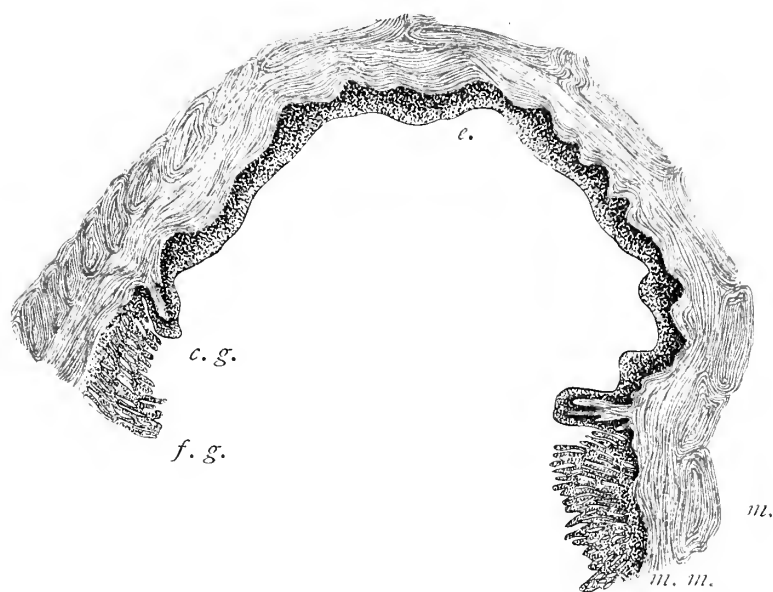


Fig. III.

Fig. IV.





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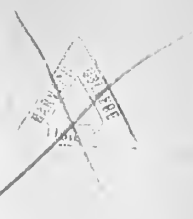
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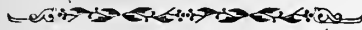
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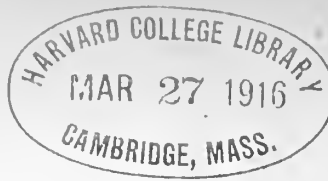
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# ON THE INHERITANCE OF THE FLOWERING TIME IN PEAS AND RICE.

By

**Yuzo Hoshino**, *Nōgakushi*.

---

## INTRODUCTION.

The multiple-factor hypothesis, which was proposed by Nilsson-Ehle (1908) for the interpretation of the inheritance of quantitative and physiological characters and carried further by Lang (1911), seems to be gaining a steady acceptance in the scientific world. The investigators who have accepted the above hypothesis support their views by the facts that the  $F_1$  is intermediate between parents; that the  $F_2$  shows a wider variation than the  $F_1$ , mostly varying within a combined range of the parents, but some producing transgressive variation; while in the  $F_3$ , some vary like the  $F_2$ , others, in several grades of variation ranges. The majority of the investigators, however, do not explain the exact number and nature of the factors which may concern the heredity of the characters.

So far as the author knows, there are only two works which touch on the latter subject. The one is Tschermak's (1911) and the other is Punnet and Bailey's (1914). Tschermak proposed, for the interpretation of his experimental results on the inheritance of the flowering time in peas, two alleromorphic pairs; Punnet and Bailey, for the interpretation of their experiments on the inheritance of weight in poultry, four alleromorphic pairs. If we examine their work carefully, however, we can not readily accept their interpretations. We shall not enter here into a minute discussion of the subject, but let us simply make the following remarks. In the work of Tschermak

mak, the grouping of the individuals in the  $F_2$  into three classes (early, intermediate and late), on the numerical ratios of which Tschermak layed great stress, seems to be quite arbitrary, and the materials of the  $F_3$  and  $F_4$  raisings are insufficient for the affirmation of the proposed interpretation. In regard to the work of Punnett and Bailey, the experimental material seems to us too scanty to prove their assumption positively.

Thus, so long as the number and nature of the factors are not definitely explained, we should have still to hold with Shull (1914), that "for the present, the hypothesis that plural Mendelian genes adequately account for the inheritance of complex quantitative and physiological character, is valuable only to the extent that it is made a working hypothesis."

The author has continued the present experiments since 1907. In the experiments with peas, he has raised the  $F_2$  and the  $F_3$  three times and the  $F_4$  twice, and, besides, he has paid special attention to the inheritance of the flowering time of pure lines in the population of parent varieties. The total number of individuals which he has raised up to the present is over 30,000. As the results of this large raising, the author feels quite confident that the present work will cast a clearer light than has hitherto been shed upon the multiple-factor hypothesis, which will be applicable to the inheritance of such a physiological character as flowering time.

While the author was conducting experiments on the inheritance of flowering time, he noticed the presence of some correlation between the flowering time and flower colour in peas, and found that this correlation might be interpreted by the assumption of gametic coupling.

The author wishes to make grateful acknowledgement for the faithful assistance of the following gentlemen;—Messrs. G. Shimada and W. Ogawa, his former assistants, and Mr. Y. Shima, his present assistant.

*Sapporo, April, 1915.*

## I. EXPERIMENTS WITH PEAS.

### Experimental Methods.

The Experiments were carried on in the College Vegetable Garden from 1907 to 1911, and in the nursery ground of the College Orchard from 1912 to 1914. As peas do not flourish under continuous cropping, care was taken to change the raising place every year, selecting even and homogeneous plots.

The method of raising the offsprings was as follows. At first, we selected a certain number of  $F_1$  plants, and took 30 seeds from each of these selected plants and sowed them according to their origin on different rows. Thus we got a certain number of  $F_2$  families, each of which consisted of 30 individuals. In raising the  $F_3$  progenies, we selected certain  $F_2$  families (rows), and took 30 seeds from every plant of these selected families, sowing them on different rows according to their different origins, just as in the case of the  $F_2$  raising.  $F_4$  raisings were also made according to the same principles.

Thus our method was to raise 30 individuals in one family and to record their flowering time, but, in reality, there were some seeds which failed to sprout, and some individuals which, on account of retarding of flowering time caused by some injuries, such as breaking of stem, insect injury etc., had to be discarded later from records. And moreover, there occurred some few individuals which flowered normally but did not produce a sufficient number of seeds for raising 30 offsprings. Such were the causes of the insufficient number of the individuals in one family that we noticed everywhere throughout the experiments. We did not raise the progenies of those plants which produced too small a number of seeds.

The rows, on which the individual plants were grown 6 inches apart, were 15 feet long and 2 feet apart. When the plants had grown about 10 inches, we gave to every plant a bamboo cane for support, binding the stem to it lightly. Afterwards we repeated this binding two or three times to

prevent entanglement with the adjoining plants. We recorded as the flowering time of every plant, the time when the standard of its first flower opened.

### Preliminary Experiments with Peas.

#### Materials Used.

When the author returned to Sapporo after four years' absence for studies abroad and took charge of the Department of Horticulture of the College, he found in the Department the stock seeds of over twenty varieties of peas which had been harvested in the previous year in the College Vegetable Garden. The dates of the flowering for all of these varieties had been recorded every year, and he could select two varieties which had a difference of about two weeks in their flowering times. A brief description of these two varieties follows.

1. Early variety ; Dwarf, white-flowered, tough-podded, white-seeded ; cultivated over ten years under the name of "Improved Dwarf" in our vegetable garden. We shall denote this variety as "**I. P.**"
2. Late variety ; Pole, red-flowered, soft-podded, gray-seeded ; cultivated longer than the former in the garden under the name of "French Large Podded". Afterwards, we identified this variety as "Pois sans parchemin jéant, à très large cosse", according to the description in Vilmorin-Andrieux' "Plantes potagères". We shall designate this variety by the abbreviation "**G. P.**".

In the early part of May, 1907, we sowed twenty seeds of **G. P.**; and when the plantlets had grown up 2 or 3 inches high, we thinned them to ten plants. After the sowing of the **G. P.** seeds, we sowed the **I. P.** seed three times at intervals of ten days. When both varieties came to flower, artificial crossing was practised between them, care being taken to record the pollen parents. When the ripened seeds were harvested, there were 54 seeds of ♀ **G. P.** × ♂ **I. P.** and 23 seeds of ♀ **I. P.** × ♂ **G. P.**

TABLE 1.—Frequency distribution of number of days from sowing to flowering in  $F_1$  of crosses between *G. P.* and *I. P.* (1908)

| Designation   | Class centers |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
|---------------|---------------|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|
|               | 43            | 44 | 45 | 46 | 47 | 48 | 49 | 50 | 51 | 52 | 53 | 54 | 55 | 56 | 57 | 58 | 59 | 60 | 61 | 62 | 63 | 64 | 65 | 66 | 67 | 68 | 69 | 70 | 71 | 72 | 73 | 74 | 75 |
| G. P.         |               |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    | 2  | 6  | 3  | 6  | 4  | 2  | 5  | 10 | 2  | 5  |    | 2  | 1  |
| I. P.         |               |    | 13 | 18 | 8  | 7  | 2  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| G. P. × I. P. |               |    |    |    |    |    |    |    |    |    |    |    |    |    |    | 2  | 6  | 7  | 14 | 7  | 2  | 6  | 2  |    | 3  |    | 1  | 1  |    |    |    |    |    |
| I. P. × G. P. |               |    |    |    |    |    |    |    |    |    |    |    |    |    | 1  | 2  |    | 4  | 2  | 4  | 5  | 2  |    | 1  | 1  |    |    |    |    |    |    |    |    |

TABLE 2.—Frequency distribution in  $F_2$  of crosses between *G. P.* and *I. P.* (1909)

| No. | Designation | Class Centers                         |       |        |        |       |       |       |        |       |       |       |        |       |        |        |        |                                       |        |        |        |        |        |        |       |       |       |       |       |    |    |    |    |    |    |    |       |    |
|-----|-------------|---------------------------------------|-------|--------|--------|-------|-------|-------|--------|-------|-------|-------|--------|-------|--------|--------|--------|---------------------------------------|--------|--------|--------|--------|--------|--------|-------|-------|-------|-------|-------|----|----|----|----|----|----|----|-------|----|
|     |             | 44                                    | 45    | 46     | 47     | 48    | 49    | 50    | 51     | 52    | 53    | 54    | 55     | 56    | 57     | 58     | 59     | 60                                    | 61     | 62     | 63     | 64     | 65     | 66     | 67    | 68    | 69    | 70    | 71    | 72 | 73 | 74 | 75 | 76 | 77 | 78 | 79    | 80 |
|     | I. P.       |                                       | 14    | 5      | 3      |       |       |       |        |       |       |       |        |       |        |        |        |                                       |        |        |        |        |        |        |       |       |       |       |       |    |    |    |    |    |    |    |       |    |
|     | I. P.       | 1                                     | 9     | 6      | 3      | 1     |       |       |        |       |       |       |        |       |        |        |        |                                       |        |        |        |        |        |        |       |       |       |       |       |    |    |    |    |    |    |    |       |    |
|     | I. P.       |                                       | 8     | 8      | 1      |       | 1     |       |        |       |       |       |        |       |        |        |        |                                       |        |        |        |        |        |        |       |       |       |       |       |    |    |    |    |    |    |    |       |    |
|     | I. P.       |                                       | 10    | 1      | 5      | 1     |       |       |        |       |       |       |        |       |        |        |        |                                       |        |        |        |        |        |        |       |       |       |       |       |    |    |    |    |    |    |    |       |    |
|     | G. P.       |                                       |       |        |        |       |       |       |        |       |       |       |        |       | 2      | 4      | 2      | 1                                     |        | 1      | 2      | 2      | 4      |        | 1     | 1     |       | 1     |       |    |    |    |    |    |    |    |       |    |
|     | G. P.       |                                       |       |        |        |       |       |       |        |       |       |       |        |       | 1      | 2      | 3      | 1                                     | 2      |        | 1      | 2      | 4      | 3      | 1     | 1     |       |       |       |    |    |    |    |    |    |    |       |    |
|     | G. P.       |                                       |       |        |        |       |       |       |        |       |       |       |        |       | 3      | 2      | 2      | 2                                     | 1      |        | 2      |        | 3      | 3      | 1     |       |       |       | 1     |    |    |    |    |    |    |    |       |    |
|     | G. P.       |                                       |       |        |        |       |       |       |        |       |       |       |        |       | 2      | 1      | 1      | 2                                     | 1      | 1      | 1      | 2      | 3      | 2      | 2     | 2     |       |       |       |    |    | 1  |    |    |    |    |       |    |
| 1   | H × 13.1    | 2                                     | 1     | 2      | 1      |       |       | 1     |        |       |       | 1     |        | 1     |        | 1      | 3      |                                       | 1      |        | 1      | 1      | 1      | 1      | 1     |       |       |       | 3     |    |    |    |    |    |    |    |       |    |
| 2   | H × 13.3    |                                       |       |        | 2      | 1     |       |       | 1      | 2     |       | 1     |        | 1     |        | 1      | 1      | 1                                     | 1      | 3      | 2      |        |        | 2      |       |       |       | 1     |       |    |    |    |    |    |    |    |       |    |
| 3   | H × 13.4    |                                       | 1     | 1      | 3      |       | 1     |       | 1      | 1     |       | 1     |        | 2     |        | 1      |        |                                       |        | 1      | 2      |        | 2      | 1      |       |       |       | 1     |       |    |    |    |    |    |    |    |       |    |
| 4   | H × 13.5    | 2                                     |       | 1      | 1      | 1     |       | 1     |        | 2     |       | 1     |        | 1     |        | 3      |        |                                       | 1      | 2      | 2      |        | 1      | 1      |       | 1     |       |       |       |    |    |    |    |    |    | 1  |       |    |
| 5   | H × 13.6    |                                       | 2     | 1      | 1      |       |       |       | 1      |       | 1     |       | 4      |       | 1      |        | 1      | 2                                     |        | 2      | 1      | 1      |        | 1      |       |       |       |       |       |    |    |    |    |    |    |    |       |    |
| 6   | 16 × 11.1   |                                       |       | 1      | 1      | 1     |       | 1     | 1      |       |       | 3     |        | 3     |        | 1      | 1      | 2                                     | 1      | 1      | 1      | 1      |        |        |       | 1     |       |       |       |    |    |    |    |    |    |    |       |    |
| 7   | 16 × 11.2   |                                       |       | 1      | 1      |       | 1     |       | 1      |       | 2     |       |        | 1     | 1      | 2      | 1      | 3                                     |        | 2      | 1      |        | 2      |        |       | 2     |       |       |       |    |    |    |    |    |    |    |       |    |
| 8   | 16 × 11.3   | 1                                     |       |        | 1      |       |       |       | 2      |       |       | 3     | 1      | 3     | 1      | 2      | 1      | 1                                     | 1      | 1      | 2      | 1      |        |        |       |       |       |       | 1     |    |    |    |    |    |    |    |       |    |
| 9   | 16 × 11.4   |                                       | 1     | 3      |        |       | 1     | 1     | 1      |       | 1     |       | 1      |       | 4      | 1      |        | 1                                     |        | 1      | 1      | 1      | 1      |        | 1     |       |       |       |       |    |    |    |    |    |    |    |       |    |
| 10  | 16 × 11.5   |                                       |       |        |        |       |       | 3     | 1      | 1     |       |       | 1      | 1     | 1      |        | 1      | 1                                     | 2      | 2      | 1      |        | 3      |        |       |       |       |       |       |    |    |    |    |    |    |    |       |    |
| 11  | V × 17.1    | 1                                     |       |        |        |       |       |       | 3      | 1     | 1     |       |        | 1     | 1      |        | 2      | 4                                     |        |        |        |        |        | 2      | 4     | 1     |       |       |       |    |    |    |    |    |    |    |       |    |
| 12  | V × 17.2    |                                       | 2     | 1      |        |       |       |       | 1      | 1     | 1     |       | 1      | 1     | 2      |        | 4      |                                       |        | 2      |        |        | 3      |        |       |       |       | 1     |       |    |    |    |    |    |    |    |       |    |
| 13  | V × 17.3    | 1                                     |       |        | 1      | 1     |       | 1     | 1      |       |       |       |        | 4     | 1      |        |        | 3                                     |        | 1      |        |        | 2      | 3      |       |       |       |       |       |    |    |    |    |    |    |    |       |    |
| 14  | V × 17.4    |                                       | 1     |        |        | 2     |       | 1     | 1      |       |       |       | 1      | 2     |        | 4      | 2      | 2                                     |        | 2      |        |        | 1      |        |       |       | 1     |       |       |    |    |    |    |    |    |    |       |    |
| 15  | 20 × V.1    | 1                                     |       |        |        | 1     | 1     | 1     |        | 1     | 1     | 3     | 1      | 1     | 1      |        | 1      | 1                                     | 1      |        | 1      |        |        |        |       |       |       |       |       |    |    |    |    |    |    |    |       |    |
| 16  | 20 × V.2    |                                       |       |        | 3      |       |       | 1     |        |       |       |       | 1      | 2     | 2      |        | 4      |                                       |        | 1      | 1      | 2      | 1      |        |       |       |       |       | 1     |    |    |    |    |    |    |    |       |    |
| 17  | 20 × V.3    |                                       | 1     |        |        |       |       | 1     |        |       |       |       | 5      |       | 3      |        | 5      |                                       |        |        |        | 1      |        |        |       |       |       |       |       | 1  |    |    |    |    |    |    |       |    |
| 18  | 20 × V.4    |                                       |       | 1      | 2      | 1     |       | 2     |        |       |       | 2     |        | 3     |        | 1      |        | 2                                     | 2      | 1      |        | 1      |        |        |       |       |       |       |       |    |    |    |    |    |    |    |       |    |
| 19  | 20 × V.5    |                                       |       | 2      |        | 2     |       | 1     |        |       | 1     |       |        | 1     | 1      | 1      | 1      | 4                                     |        | 1      | 2      | 1      |        | 3      |       |       |       |       |       |    |    |    |    |    |    |    | 0 : 1 |    |
|     |             | 4 : 4                                 | 7 : 0 | 11 : 4 | 12 : 2 | 5 : 3 | 4 : 0 | 6 : 5 | 6 : 12 | 2 : 7 | 0 : 7 | 1 : 4 | 1 : 23 | 0 : 8 | 3 : 36 | 1 : 12 | 1 : 15 | 2 : 42                                | 0 : 10 | 1 : 14 | 1 : 22 | 0 : 13 | 0 : 17 | 0 : 19 | 1 : 5 | 0 : 1 | 4 : 0 | 0 : 3 | 0 : 0 |    |    |    |    |    |    |    |       |    |
|     |             | 61 + 48 = 112 (Early flowering group) |       |        |        |       |       |       |        |       |       |       |        |       |        |        |        | 12 + 247 = 259 (Late flowering group) |        |        |        |        |        |        |       |       |       |       |       |    |    |    |    |    |    |    |       |    |

Black-faced type indicates number of white-flowered individuals.  
 indicates number of dwarf individuals.

TABLE 3.—Frequency distribution in  $F_2$  of crosses between *G. P.* and *I. P.* (1910)

| No. | Designation  | Parent | Class Centers |    |    |     |     |      |     |    |     |    |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     | Means of Constants |    |    |    |    |       |           |           |
|-----|--------------|--------|---------------|----|----|-----|-----|------|-----|----|-----|----|----|-----|----|-----|----|-----|----|----|-----|-----|----|-----|----|----|-----|--------------------|----|----|----|----|-------|-----------|-----------|
|     |              |        | 46            | 47 | 48 | 49  | 50  | 51   | 52  | 53 | 54  | 55 | 56 | 57  | 58 | 59  | 60 | 61  | 62 | 63 | 64  | 65  | 66 | 67  | 68 | 69 | 70  |                    | 71 | 72 | 73 | 74 | 75    |           |           |
|     | I. P.        |        |               | 1  | 5  | 17  | 23  | 4    | 6   | 1  | 1   |    |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    | 40.88 |           |           |
|     | G. P.        |        |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    |     |    |    | 2   | 20  | 31 | 18  | 4  | 6  | 2   | 1                  |    | 2  |    |    | 67.55 |           |           |
| 1   | V × 17.2.12  | 46     |               |    | 1  | 8   | 18  | 3    |     |    |     |    |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    | 40.77 |           |           |
| 2   | V × 17.2.17  | 46     |               |    | 5  | 8   | 11  | 1    | 1   | 1  |     |    |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    | 49.50 |           |           |
| 3   | H × 13.1.9   | 45     |               |    | 1  | 3   | 17  | 3    | 3   |    |     |    |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    | 50.35 |           |           |
| 4   | V × 17.1.2   | 45     |               |    | 1  | 5.4 | 9.2 | 2    |     |    |     |    |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    | 49.01 |           |           |
| 5   | 16 × 11.2.14 | 47     |               |    |    | 2   | 4   | 10   | 4   | 5  |     | 2  |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    | 51.52 |           |           |
| 6   | 16 × 11.2.29 | 47     |               |    |    | 3.1 | 3.3 | 3    | 9.4 | 4  | 1   |    |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    | 51.60 |           |           |
| 7   | 16 × 11.1.3  | 47     |               |    |    | 3   | 8   | 3    | 8   | 3  |     |    |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    |       | 51.60     |           |
| 8   | 16 × 11.1.17 | 48     |               |    |    | 1   | 10  | 2    | 7   | 5  | 3   |    |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    |       | 51.50     |           |
| 9   | H × 13.1.21  | 47     |               |    | 1  |     | 6   | 6    | 8   | 5  |     |    |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    |       | 51.35     |           |
| 10  | V × 17.2.4   | 47     |               |    |    |     | 8   | 4    | 5   | 2  |     |    |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    |       | 51.05     |           |
| 11  | H × 13.1.9   | 46     |               |    |    |     | 1   | 7    | 11  | 2  | 3   |    |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    |       | 51.60     |           |
| 12  | 16 × 11.1.20 | 48     |               |    |    | 1   | 2   | 10.3 | 4   | 4  | 5.2 |    |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    |       | 51.93     |           |
| 13  | H × 13.3.2   | 49     |               |    |    | 3   | 2   | 3    | 5   | 3  | 11  |    |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    |       | 52.33     |           |
| 14  | 16 × 11.2.22 | 49     |               |    |    |     |     | 1    | 9   | 6  | 3   | 1  |    |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    |       | 53.70     |           |
| 15  | 20 × V.1.1   | 49     |               |    | 2  | 1   | 1   | 2    |     | 6  | 4   | 1  | 4  |     |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    |       | av. 51.19 |           |
| 16  | 20 × V.2.19  | 48     |               |    | 1  | 1   |     | 1    | 6   | 6  | 1   | 4  | 5  | 2   |    |     |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    |       |           |           |
| 17  | H × 13.3.7   | 48     |               |    |    |     |     | 1    | 5   | 6  | 2   | 1  | 3  | 1   | 2  | 3   |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    |       |           |           |
| 18  | H × 13.1.19  | 47     |               |    |    |     |     | 1    | 5   | 6  | 2   | 1  | 1  | 1   | 2  | 5   | 5  |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    |       |           |           |
| 19  | H × 13.3.19  | 48     |               |    |    |     |     | 1    | 5   | 6  | 2   | 1  | 3  | 1   | 3  | 3   |    |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    |       |           |           |
| 20  | 20 × V.2.7   | 48     |               |    |    |     |     | 2    | 1   |    | 7   | 2  | 1  | 5   | 5  | 4   | 1  |     |    |    |     |     |    |     |    |    |     |                    |    |    |    |    |       |           |           |
| 21  | 16 × 11.1.2  | 63     |               |    | 1  |     |     | 2    | 13  | 2  |     |    |    |     | 1  | 1.1 | 1  | 2   |    | 2  | 2   | 1   | 1  | 5   | 2  | 1  | 1   |                    |    |    |    |    |       |           |           |
| 22  | 20 × V.2.14  | 66     |               |    |    |     | 1   |      |     |    | 5   | 1  | 1  | 1.1 |    |     |    | 2   | 2  |    | 1   | 5.1 | 2  | 5   |    |    | 1   | 1                  |    |    |    |    |       |           |           |
| 23  | H × 13.3.9   | 65     |               |    |    |     |     |      |     |    | 13  |    |    | 1   |    |     |    | 1   | 2  | 1  | 2   | 2   | 4  |     |    | 1  | 9.1 |                    |    |    |    |    |       |           |           |
| 24  | H × 13.1.12  | 68     |               |    |    |     |     |      | 1   |    |     | 1  | 1  | 1   | 1  |     |    |     | 2  | 2  | 3   | 2   | 1  | 1   |    |    |     |                    |    |    |    |    |       |           |           |
| 25  | V × 17.1.10  | 66     |               |    |    |     |     |      |     | 2  |     |    | 2  |     |    |     |    |     | 1  | 1  | 1   | 1   | 1  | 4   | 3  | 1  | 3   | 4                  | 1  |    |    |    |       |           |           |
| 26  | V × 17.2.2   | 64     |               |    |    |     |     |      |     |    |     | 1  |    | 1   |    | 1   |    | 1.1 | 1  | 1  | 3.2 | 2.1 | 0  | 3.1 | 2  | 1  | 1   | 1                  |    |    |    |    |       |           |           |
| 27  | V × 17.1.7   | 67     |               |    |    |     |     |      |     |    |     |    |    | 2   |    |     |    | 1   | 1  |    |     | 4   | 2  | 1   |    |    | 1   | 1                  |    |    |    |    |       |           |           |
| 28  | V × 17.1.19  | 68     |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    | 1   | 1  | 1  | 1   | 5   | 3  | 5   | 1  |    | 1   | 1                  |    |    |    |    |       |           |           |
| 29  | H × 13.3.14  | 64     |               |    |    |     |     |      |     |    |     | 1  |    | 2   |    | 1   | 1  |     | 1  | 1  | 3   | 4   | 2  | 4   | 4  |    | 2   | 1                  |    |    |    |    |       |           |           |
| 30  | H × 13.3.21  | 64     |               |    |    |     |     |      |     |    |     |    | 2  |     | 1  | 2   |    |     |    | 2  | 1   | 2   | 3  | 5   | 3  | 2  |     |                    |    |    |    |    |       |           |           |
| 31  | V × 17.2.14  | 66     |               |    |    |     |     |      |     |    |     | 2  |    |     |    | 2   |    |     |    |    |     |     | 3  | 3   | 1  | 1  | 2   |                    |    |    |    |    |       |           |           |
| 32  | 16 × 11.1.16 | 64     |               |    |    |     |     |      |     |    |     |    |    |     |    | 1   | 1  | 1   | 1  |    |     | 2   | 2  | 10  | 1  | 3  | 2   | 2                  |    |    |    |    |       |           |           |
| 33  | H × 13.3.10  | 71     |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    | 2   | 2  | 4  |     | 3   | 2  | 3   | 1  |    |     |                    |    |    |    |    |       |           |           |
| 34  | H × 13.5.12  | 68     |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    | 1   | 1  | 1  |     |     | 4  |     | 4  |    | 4   | 7                  | 3  |    |    |    |       |           |           |
| 35  | V × 17.2.10  | 66     |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    |     |    |    |     | 4   | 1  | 1   | 4  | 3  | 1   | 2                  | 1  | 1  |    |    |       |           |           |
| 36  | 16 × 11.1.14 | 65     |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    |     |    |    |     | 3   | 3  | 5   | 2  | 2  | 3   | 2                  |    |    |    |    |       |           | 67.70     |
| 37  | H × 13.1.22  | 66     |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    |     |    |    | 1   | 2   | 6  | 4   | 0  | 3  | 2   |                    | 1  |    |    |    |       |           | 67.86     |
| 38  | H × 13.1.9   | 65     |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    |     |    |    |     | 1   | 3  | 4   | 1  | 2  | 3   | 5                  | 2  |    |    |    |       |           | 67.60     |
| 39  | V × 17.2.8   | 64     |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    |     |    |    |     | 2   | 1  | 6   |    | 5  | 10  | 2                  |    |    |    |    |       |           | 67.71     |
| 40  | 16 × 11.2.6  | 67     |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    |     |    |    |     |     | 3  |     |    |    |     | 1                  | 1  | 3  |    |    |       |           | 68.13     |
| 41  | 20 × V.2.8   | 72     |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    |     |    |    |     |     |    |     |    | 2  | 3   | 1                  | 1  | 2  | 1  |    |       |           | 68.04     |
| 42  | H × 13.1.7   | 72     |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    |     |    |    |     |     |    |     |    | 2  | 5   | 10                 | 4  | 3  |    |    |       |           | 68.56     |
| 43  | H × 13.3.11  | 68     |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    |     |    |    |     |     |    |     | 1  | 3  | 3   | 8.56               | 5  | 1  | 1  |    |       |           | 69.10     |
| 44  | 20 × V.2.16  | 65     |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    |     |    |    |     |     |    |     |    | 2  | 2   | 2                  | 4  | 1  |    |    |       |           | 69.72     |
| 45  | 20 × V.2.5   | 67     |               |    |    |     |     |      |     |    |     |    |    |     |    |     |    |     |    |    |     |     |    |     | 1  | 2  | 5   | 5                  | 3  | 2  |    |    |       |           | av. 68.10 |





**F<sub>1</sub> Raising in 1908.**

On May 8, we sowed all of the cross seeds harvested in the previous year, together with some seeds from the parent plants which were used in the crossing as the seed parents or the pollen parents. With a few exceptions, the seeds sprouted very evenly, and the young plants grew in excellent condition.

During the summer vacation of that year, the author had to take a trip to Manchuria, and at the time when the **I. P.** plants had just begun to flower, he left Sapporo, entrusting all observations to his assistant. On his return to Sapporo in the middle of September, he found all had been harvested in rather miserable condition, and heard that in August a formidable storm had raged in Northern Japan, and many of our plants were badly injured and many of the ripened pods were shaken off. And to our great regret, owing to the misunderstanding of the assistant, all the pods of every parent variety were sacked together without any distinction of the origin.

But, from the records taken, we got the frequency distribution of the number of days from sowing to flowering of the **F<sub>1</sub>** offsprings and that of the parent varieties, as shown in Table 1.

From Table 1, we see that the flowering time of the **F<sub>1</sub>** is not just intermediate between those of the parent varieties, but inclines much towards that of the late parent. In other words, we may say that in the present instance incomplete dominance of lateness occurs. And there is practically no essential difference between reciprocal crosses.

**F<sub>2</sub> Raising in 1909.**

From the **F<sub>1</sub>** plants we selected the following individuals:—

- |                   |   |                    |        |       |       |       |       |
|-------------------|---|--------------------|--------|-------|-------|-------|-------|
| ♀ <b>G. P.</b> 11 | × | ♂ <b>I. P.</b> 13, | —No. 1 | No. 3 | No. 4 | No. 5 | No. 6 |
| ♀ <b>I. P.</b> 16 | × | ♂ <b>G. P.</b> 11, | —No. 1 | No. 2 | No. 3 | No. 4 | No. 5 |
| ♀ <b>G. P.</b> 5  | × | ♂ <b>I. P.</b> 17, | —No. 1 | No. 2 | No. 3 | No. 4 |       |
| ♀ <b>I. P.</b> 20 | × | ♂ <b>G. P.</b> 5,  | —No. 1 | No. 2 | No. 3 | No. 4 | No. 5 |

Notice: Hereafter, we shall denote the plants of a late parent by Roman numerals and those of an early parent by arabic figures, and in the crosses, the antecedent will be the seed parent, the subsequent, the pollen parent. For example,  $\text{II} \times 10$  will denote the product of the No. 2 plant of the late parent variety crossed with the No. 10 plant of the early parent variety.

We took 20 seeds from each of the selected 19  $F_1$  individuals and sowed them on May 6, together with 80 seeds (in 4 rows) of each parent varieties. Sprouting took place evenly, during the two days May 14, 15.

The frequency distribution of the number of days from sowing to flowering in the  $F_2$  and parent varieties is shown in Table 2. Reserving the discussion of the results of the present experiments for a later chapter, we shall simply enumerate here the chief facts observed.

1. The variation ranges of the  $F_2$  families extend from the range of the early parent to that of the late parent. But there does not appear any transgressive variation. In the family  $\text{II} \times 13.5$ , however, there is one individual whose flowering date exceeds the variation range of the late parent; and it was an exceedingly dwarf (12.5 inches) and weak plant. This abnormal retarding of the flowering time might have been caused by insufficient light and nutrition, as it grew amidst the other stouter and larger plants.

2. By summing up the frequencies of each class perpendicularly in the table, we see a class which has a comparatively small number of frequencies situated at about the middle of the variation means of both parent varieties (class 55). As this fact has a great bearing on our experiments throughout, we shall designate this class as the *minimum frequency class*. The individuals which flowered before this class (this class inclusive) shall be considered as belonging to the *early flowering group*, those which flowered after the class, as belonging to the *late flowering group*.

3. In the early flowering group the white-flowered individuals are more numerous than the red-flowered ones, and in the late flowering group the white-flowered individuals are exceedingly few, and indeed fewer than the red-

flowered ones in the early group. This suggests some correlation between the flower color and the flowering time.

4. The ratios of poles to dwarfs is approximately 3 : 1 in total, and when considered in two separate groups, the ratio also holds the same in each. So we may conclude that there is no correlation between the stem character and the flowering period.

5. We do not see any difference between the progenies of reciprocal crosses.

6. The variation range of the progeny of the late variety **G. P.**, is distinctly wider than that of the **I. P.** progeny. We had noticed a similar fact already in the previous year, but did not pay much attention to it. And the frequency distribution of **G. P.** was quite different from that in ordinary fluctuating variation. In the ordinary variation, the mode must lie in about the middle of the range, and towards both sides of the mode the frequencies must decrease gradually, but in this case, the frequencies in the class 64, situated in about the middle of the variation range, are remarkably small, while in the classes 60 and 67, the frequencies are prominently large. Why did such an irregularity in the variation type occur? This might occur by chance in such a case as this, where only 80 individuals were observed, but it can not be denied that there may be some other complicated causes for such an occurrence. For example, if the population of **G. P.** should consist of from more than one pure line, or if the character of the flowering time were not fixed and if hereditary variation should to some extent occur, there would appear such a variation type as mentioned above. Thus, we were quite convinced that unless we raised hybrid progenies together with the progenies of their original parents and the hereditary modes of the parent varieties themselves are elucidated, we could not reach any exact conclusions. But because of careless harvesting of the seeds of the parent plants in the previous year, we were not able to distinguish the origin of the offsprings of the parent varieties in the present year.

Besides, we found that **I. P.** was not an adequate parent variety for the

experiment, because the flowering time of the dwarf plants which appear as the result of segregation may be influenced by the shading and usurpation of the nutrient of larger growing sister plants, and some of those dwarfs would not produce sufficient seeds for further experiments. (One individual class 79 of the family  $\text{II} \times 13.5$ , already referred to, produced only 3 pods and 8 seeds)

From the two above mentioned causes, we determined to take new crosses and to conduct the experiments anew.

### **F<sub>3</sub> Raising in 1910.**

Having determined to continue the experiments with new materials, we made new crosses during February and March of the present year in a forcing house. But as there was some space in the garden available for the experiment, we raised the  $F_3$  for the purpose of using the results of this raising for reference in our new experiments.

We selected 20  $F_2$  individuals which flowered within the variation range of the **I. P.** variety, 25  $F_2$  individuals which flowered within the variation range of **G. P.** variety, 3 **G. P.** individuals which flowered in 67 days, and 3 **I. P.** individuals which flowered in 46 days. We tried to take 30 seeds from each of the selected individuals, but some dwarf individuals did not produce the required number of seeds, as in the case of the parents of the last two families No. 44 and 45 in Table 3.

The sowing of the seeds was done on May 5. After the sowing, dry weather prevailed and sprouting was retarded, but on the 23rd and 24th of that month, plantlets appeared evenly.

The results are shown in Table 3. From this table we see that some of the selected early and late  $F_2$  plants produced offsprings whose variation ranges and types strongly suggest that the parent  $F_2$  plants had become constant (homozygous) in their flowering character (Nos. 1-14 in the early; Nos. 36-45 in the late), and the remaining  $F_2$  plants produced offsprings whose variation types are irregular and suggest that they must be offsprings of

variable (heterozygous) parents (Nos. 15-20 in the early; Nos. 21-35 in the late). The variation ranges and means of the would-be constant families descended from the early or the late  $F_2$  are not in exact coincidence with those of the corresponding parent varieties. For example, the variation means of the families No. 1 to No. 4 are very near to, but those of the families No. 5 to No. 14 are larger than, those of **I. P.**, especially that of No. 14. And in the variation types of the variable families, there is much irregularity and complicacy; for example, in the variable families from the selected late  $F_2$ , No. 21 and No. 22 vary with the combined ranges of both parent varieties, while other variable families vary with different narrower widths of range.

Thus we see, as Tschermak (1904) already observed, that among those  $F_2$  plants which flowered within the variation range of the early parent variety, as well as among those which flowered within the variation range of the late parent variety, only one part was constant and the other part showed segregation. As to the interpretation of such complicated phenomena, we shall treat it fully later in the present paper.

### Experiments Proper with Peas.

For the reason stated in the preceeding pages, we selected a new variety for the early parent in the present experiments. A short description of the variety is as follows:—

Half dwarf, 86.02 cm. high (measured in 1910); white-flowered; tough-podded; flowering time about the same as **I. P.**; cultivated for over 10 years in the College Vegetable Garden under the name "Mans". We shall denote this variety as "**M. P.**".

We selected some 50 seeds of both **G. P.** and **M. P.** from the stock seeds harvested in 1909 in the sample section of the Garden, and sowed 32 seeds of **G. P.** on Dec. 2, 1909, in a forcing house. From the 9th to 11th of the same month, all the seeds sprouted, and after the plantlets grew to about 2 inches high, one half of them were thinned out. The first sowing of

the **M. P.** seeds (16 in number) was made on Jan. 11, 1910, and the second on the 25th of the same month. From our experience in the raising of the  $F_2$  of the **G. P.**  $\times$  **I. P.** cross in the previous winter, we found the above mentioned intervals in the sowing times of the parent varieties to be suitable for crossing. The thinning of **M. P.** was done in the same manner as in the case of **G. P.**

The dates on which first flowers appeared were as follows :—

| <b>G. P.</b> |       |         | <b>M. P.</b>     |      |         |
|--------------|-------|---------|------------------|------|---------|
| No.          | I     | Feb. 19 | No.              | 1    | Feb. 21 |
| " XV         | "     | 19      | First<br>Sowing  | " 2  | " 24    |
| " II         | "     | 20      |                  | " 3  | " 24    |
| " XI         | "     | 20      |                  | " 4  | " 22    |
| " III        | "     | 24      |                  | " 5  | " 24    |
| " X          | "     | 24      |                  | " 6  | " 23    |
| " XVI        | "     | 24      |                  | " 7  | " 24    |
| " VII        | "     | 25      | Second<br>Sowing | " 8  | " 24    |
| " IV         | March | 7       |                  | " 9  | March 8 |
| " XII        | "     | 8       |                  | " 10 | " 9     |
| " XIV        | "     | 10      |                  | " 11 | " 9     |
| " VI         | "     | 11      |                  | " 12 | " 9     |
| " V          | "     | 12      |                  | " 13 | " 8     |
| " XIII       | "     | 14      |                  | " 14 | " 11    |
| " VIII       | "     | 18      |                  | " 15 | " 9     |
| " IX         | "     | 20      |                  | " 16 | " 9     |

Here we see again that the variation range of the early variety is quite narrow, while that of the late variety is remarkably wide.

By crossing two varieties, we got 106 seeds in ♀ **G. P.**  $\times$  ♂ **M. P.** and 64 seeds in ♀ **M. P.**  $\times$  ♂ **G. P.** The cross seeds reached maturity suitable for sowing in the middle of May. So we sowed these cross products together with some seeds of both parents on May 21.

Before recording the flowering times of the hybrid progenies, we shall first treat here the observations on the inheritance of the flowering time in

the parent varieties.

## Inheritance of the Flowering Time in the Progenies of the Parent Varieties.

### Raising in 1910.

From every plant which was used as a parent of the cross in the forcing house, we collected all pods which had been left untouched and contained self-fertilized seeds. As mentioned above, the sowing of those seeds together with the cross products was done on May 21. Besides, we took 100 seeds of **G. P.** and 50 seeds of **M. P.** from the stock seeds of the Vegetable Garden and sowed them at the same time.

After the sowing of the seeds, dry weather continued, and they germinated unevenly. The first sprouting appeared in May 30, and it continued until the middle of June. In this case, we can not take simply the number of days from sprouting to flowering as the representative value of the flowering time, because the late-sprouted individuals have always the tendency to hasten their flowering on account of a comparatively high temperature. In fact, by examining the flowering time of every individual of the **M. P.** variety in whose offspring we should expect a narrow range and ordinary type of the variation, from the results obtained in the raising of the previous winter, we see that the number of days from sprouting to flowering was always smaller in the later-germinated individuals than in the earlier-sprouted ones. So we adapt the following formula for finding a representative value of the flowering time :

$$\frac{\text{Number of days from sowing to flowering} + \text{that from sprouting to flowering.}}{2}$$

By calculating the representative values of all **M. P.** individuals by this formula, and arranging them in a frequency distribution table, we obtained the result which is shown in Table 4. (The designation **L** means a family consisting of 50 plants grown from the stock seeds). In the table, we see small ranges and regular types of variation in all families of the **M. P.** offsprings,

as we had expected.

The frequency distribution of representative values calculated by the above mentioned formula in the **G. P.** offsprings, is shown in the same table. (The designation **C** means a family of 100 plants grown from the stock seeds) Here, we see much irregularity of variation. The offsprings of those plants which flowered early in the forcing house (I. XV. XI. XVI.) flowered distinctly earlier than those of the late-flowered plants (XIV. VI. VIII.). In the family **C**, the variation range is quite wide and there are two distinct classes with a larger frequency (class 50 and 55).

#### Raising in 1911.

From the plants grown in the previous year, we selected the following individuals:—

|    |             |      |              |          |
|----|-------------|------|--------------|----------|
| 2  | individuals | from | <b>M. P.</b> | I        |
| "  | "           | "    | "            | 6        |
| "  | "           | "    | "            | 12       |
| 3  | "           | "    | <b>G. P.</b> | I        |
| "  | "           | "    | "            | XII      |
| "  | "           | "    | "            | VI       |
| "  | "           | "    | "            | VIII     |
| 20 | "           | "    | "            | <b>C</b> |

In selecting these individuals from each family, we took care to choose those which flowered on different dates and which made normal growth. We sowed 30 seeds from each selected individual on May 6. After the sowing, extraordinarily dry weather followed, and during the 3 days from the 15th to the 17th of the month, only a partial sprouting occurred, and afterwards the sprouting stopped almost entirely. So, we were obliged to water artificially on the ungerminated parts. From the 26th of the month they were watered every day. As a consequence, sprouting began to take place from the 31st of the month and continued until the 4th of June.

Here, again, we can not use simply the number of days from sowing to flowering as a representative value of the flowering time. The mode of



TABLE 4.—*Frequency distribution of representative value in parent varieties (1910)*

| Designation | Class centers |    |    |    |    |    |    |    |    |    | Designation        | Class centers |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
|-------------|---------------|----|----|----|----|----|----|----|----|----|--------------------|---------------|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|
|             | 30            | 31 | 32 | 33 | 34 | 35 | 36 | 37 | 38 | 39 |                    | 46            | 47 | 48 | 49 | 50 | 51 | 52 | 53 | 54 | 55 | 56 | 57 | 58 | 59 | 60 | 61 | 62 | 63 | 64 | 65 |
| M. P. 1     |               |    | 1  | 6  | 1  |    |    |    |    |    | G. P. I (19. Feb.) |               |    | 2  | 2  | 4  | 3  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| " 4         |               |    | 1  | 2  | 3  |    |    |    |    |    | " XV ( " " )       |               |    | 1  | 2  |    | 1  | 3  | 1  | 1  |    |    |    |    |    |    |    |    |    |    |    |
| " 5         |               |    | 2  | 7  | 1  | 1  |    |    |    |    | " XI (20. " )      |               |    | 1  | 2  | 3  | 1  |    | 1  |    | 1  |    |    |    |    |    |    |    |    |    |    |
| " 6         |               | 1  | 1  | 3  | 2  | 5  | 1  |    |    |    | " X (24. " )       |               |    |    | 5  | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| " 8         |               |    |    | 8  | 5  | 1  |    |    |    |    | " XVI ( " " )      |               |    | 3  | 1  | 3  | 2  | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |
| " 10        |               |    |    | 1  | 1  | 5  | 1  |    |    |    | " IV ( " March)    |               |    |    | 1  | 1  |    | 2  | 1  |    |    |    |    |    |    |    |    |    |    |    |    |
| " 11        |               |    | 1  | 3  | 4  | 6  | 1  | 1  | 1  |    | " XII (8. " )      |               |    |    | 1  | 2  | 2  | 4  | 4  | 1  | 1  |    |    |    |    |    |    |    |    |    |    |
| " 12        |               |    |    | 3  | 5  | 2  | 3  |    |    |    | " XIV (10. " )     |               |    |    |    |    |    |    |    | 4  | 2  | 1  | 2  | 1  |    |    |    |    |    |    |    |
| " 14        |               |    |    | 5  | 5  | 2  |    |    |    |    | " VI (11. " )      |               |    |    |    |    |    |    |    | 2  |    | 2  | 5  |    |    |    |    | 1  |    |    |    |
| " 15        |               |    |    | 3  | 3  | 1  |    |    |    |    | " VIII (18. " )    |               |    |    |    |    |    |    |    |    |    |    |    | 3  |    |    | 2  |    |    |    |    |
| " 1         |               | 1  | 1  | 5  | 23 | 6  | 4  | 3  |    |    | " C                |               | 1  |    | 4  | 10 | 9  | 3  | 6  | 7  | 13 | 12 | 6  | 6  | 6  | 5  | 1  |    | 1  |    |    |

TABLE 5.—*Frequency distribution in parent varieties (1911)*

Black-faced type indicates early sprouted.

TABLE 6.—Frequency distribution in different pure lines of G. P. (1912 and 1913)

<sup>c</sup> ( ) Early sprouted



sprouting in this year was different from that in the previous year. The sprouting then occurred gradually. In this year, however, we can divide all individuals into two groups, one consisting of those which sprouted during the 3 days from the 15th to the 17th of May, and the other, those which sprouted from the 31st of May to the 4th of June. In Table 5, the frequency distribution of the number of days from sowing to flowering in the two groups is shown. The black-faced types indicate the frequencies of the early-sprouted group.

From Table 5, we can affirm the following facts:—

1. The variation types of all families descending from **M. P.** are alike and uniform.
2. Among the families descending from **G. P.** there are two distinctly different types of variation. All families from I, XII and XVI show the early type of variation, and those from VI and VIII the late type of variation. Among the families from **C.**, the first 12 families are of the early type and the remaining 8 families of the late type.
3. In raising the offsprings of one common grand-parent, we took special care to select those parents plant which differed in their flowering times; and in raising those of **C** family, we selected those parents which flowered in different grades within the variation range of the family. But all families descending from a common grand-parent showed the same type of variation; and the variation types of all families from **C** fell into two distinct categories.

From the facts cited above, it can safely be concluded that the character of the flowering time in peas is a fixed and stable one, and that the population of the **G. P.** variety contains two different pure lines. By assuming the presence of two pure lines in the **G. P.** population, we can easily explain the irregularity and wide range of the variation in the **G. P.** offsprings which we have seen in several previous raisings (Tables 1, 2, 3 and 4).

### Raisings of **G. P.** Offsprings in 1912 and 1913.

Though we were quite convinced, from the previous experiments, of the stability of the flowering character, we took care, in the raisings of the **G. P.** progenies for the purpose of comparison with the hybrid progenies, to select those parent plants which differed in flowering times. In Table 6, we show the results of the raisings in 1912 and 1913, in which the stability of the flowering character and the existence of two pure lines are clearly shown.

### Inheritance of the Flowering Time in Hybrid Progenies.

#### $F_1$ Raising in 1910.

The sowing of the hybrid seeds (106 of **G. P.**  $\times$  **M. P.** and 64 of **M. P.**  $\times$  **G. P.**; Page 238) was done on the 21st of May. The sprouting of the seeds was so uneven, as to make it necessary to calculate the value of the flowering time of every individual by the formula already mentioned ( page 239).

The frequency distribution of the value of the flowering time is as shown in Table 7. From this table, we see the incomplete dominance of lateness in the  $F_1$ , and no difference between the reciprocal crosses.

It was not then known yet that there were two distinct pure lines in **G. P.** population; but as the line of every parent plant was ascertained by the later experiments, we have arranged, in the table, all families into two groups, the early and late lines. By calculating the variation means of both groups separately, we have 47.8 days in the early line group and 49.5 days in the late line group. So the variation mean of the  $F_1$  individuals whose **G. P.** parents belonged to the early pure line, is smaller than that of the  $F_1$  individuals whose **G. P.** parents belonged to the late pure line. This difference of the means in the two groups suggests that the pure line character is inheritable in itself.

As to the morphological characters of the  $F_1$  plants, the color of all of the flowers was red; the height of the stem was not just intermediate, but rather inclined towards, that of **G. P.**, the average height of 100  $F_1$  plants being 158.8 cm., while that of **G. P.** and **M. P.** was 184.48 cm. and 86.02 cm.

respectively; and the tough-poddedness seemed apparently to dominate over the soft-, but by measurement we could ascertain that the thickness of the scleren-chymatous tissue of the  $F_1$  plant was intermediate, being  $81\ \mu$ , while in **G. P.** and **M. P.** it was  $51\ \mu$  and  $100\ \mu$  respectively <sup>1)</sup>.

### $F_2$ Raisings.

#### In 1911.

From the  $F_1$  plants of the previous year, the following individuals were selected:—

|                  |                    |
|------------------|--------------------|
| I $\times$ 1.2   | VIII $\times$ 12.1 |
| I $\times$ 1.3   | III $\times$ 12.4  |
| I $\times$ 1.4   | VIII $\times$ 12.5 |
| 4 $\times$ XVI.2 | 12 $\times$ VIII.1 |
| 4 $\times$ XVI.4 | 13 $\times$ VIII.2 |
| 4 $\times$ XVI.6 |                    |

30 seeds from each selected individual were sown on May 6, but, on account of the extraordinary dryness of the weather, uneven sprouting was caused (page 240). In Table 8, group **A** includes the offsprings which had for one of their parents an early pure line individual of **G. P.**, and group **B**, a late pure line individual of the same variety. Each of the groups **A** and **B** is differentiated into two sub-groups, according to the earliness or lateness of the sprouting.

#### In 1912.

As the sprouting of the  $F_2$  plants in 1911 had been too uneven, we renewed the raising in order to obtain the variation type in the  $F_2$  with normal sprouting. For this purpose, we raised the progenies of the following  $F_1$  plants.

|                  |                  |
|------------------|------------------|
| XVI $\times$ 3.2 | 3 $\times$ XVI.1 |
| XVI $\times$ 3.3 | 3 $\times$ XVI.2 |
| XVI $\times$ 3.4 |                  |

1) For these measurements the author is indebted to Mr. R. Matsubayashi.

To our regret, no seeds of the  $F_1$  plants which were crosses of the late pure line of **G. P.** had been kept. Sown on May 7, sprouting took place evenly during the three days from the 17th to the 19th of the same month. The frequency distribution of the number of days from sowing to flowering in this raising is as shown in Table 8 **C**.

#### Chief Results Obtained from the Above Two Raisings of $F_2$

1. By summing up the frequencies of every class perpendicularly in every group of Table 8 and comparing the sums thus obtained with one another, we find in **A<sub>1</sub>**, **B<sub>2</sub>** and **C**, the minimum frequency classes (page 234) in the middle between the variation ranges of the parent varieties, 50 in **A<sub>1</sub>**, 65 in **B<sub>2</sub>** and 53 in **C**. In **A<sub>2</sub>** and **B<sub>1</sub>** groups, on account of the small number of individuals, it is rather difficult to locate this class, but judging from the position of the class in **A<sub>1</sub>** and **B<sub>2</sub>**, and comparing actual number of frequencies in every class, we may safely fix this class in **A<sub>2</sub>** at 62, and in **B<sub>1</sub>** at 52. Thus, we see that the values of the minimum frequency classes of **A<sub>1</sub>** and **A<sub>2</sub>** are smaller than those of **B<sub>1</sub>** and **B<sub>2</sub>**. That is to say, the minimum frequency class in the variation of the families descended from the crosses with the early pure line of **G. P.** is situated earlier than that of the families descended from the crosses with the late pure line of **G. P.** This indicates that the pure line character is hereditary. Now, if we examine again Table 2, we may conclude that **G. P.** II and **G. P.** V belonged to the same pure line (probably to the late flowering pure line), because we do not recognize there any difference in the location of the minimum frequency classes among the families of II  $\times$  13, 16  $\times$  II and V  $\times$  17, 20  $\times$  V.
2. When we divide all individuals of every family into two groups, the early flowering and the late flowering, taking the minimum frequency class as the demarkation line, we see that in the early group the white-flowered individuals are more numerous than the red-flowered ones, and in the late group the number of the white-flowered ones is remarkably small.

TABLE 7.—Frequency distribution of representative value in  $F_1$  of crosses between G. P. and M. P.

TABLE 8.—Frequency distribution in  $F_2$  of crosses between G. P. and M. P.

[illegible]

Black-faced type indicates number of white-flowered individuals.

2

5

1

c

f

1



This fact coincides with what we have seen in Table 2.

3. Keeble and Pellew (1910) from their experiments, concluded that there might be a gametic coupling between stem characters and flowering character. In the present experiments, we could not find the existence of such a correlation. In general, the later the flowering, the thicker the stem and the longer the internode. For example, by exact measurement of the length of the internode of every  $F_2$  individual of the raising in 1912, taking those which appeared to be representative in the length of the internodes in the individual plant, we got the following results:—

|                                                  |                                                                                                                                                                                                                                                                                                                                                                                                                   |
|--------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Length of internode of <b>M. P.</b> plants       | 6.0—7.5 cm.                                                                                                                                                                                                                                                                                                                                                                                                       |
| „ „ „ „ <b>G. P.</b> plants                      | 9.5—11.0 cm.                                                                                                                                                                                                                                                                                                                                                                                                      |
| „ „ „ of $F_2$ plants in<br>classes 46, 47 & 48. | <div style="display: inline-block; vertical-align: middle;"> <div style="display: inline-block; vertical-align: middle;"> <div style="display: inline-block; vertical-align: middle;">none shorter than 7.5 cm.</div> <div style="display: inline-block; vertical-align: middle;">none longer than 9.5 cm.</div> <div style="display: inline-block; vertical-align: middle;">majority 9.0 cm.</div> </div> </div> |
| „ „ „ of $F_2$ plants in<br>classes 49, 50 & 51. | <div style="display: inline-block; vertical-align: middle;"> <div style="display: inline-block; vertical-align: middle;">none shorter than 8.5 cm.</div> <div style="display: inline-block; vertical-align: middle;">majority 9.0 cm.</div> </div>                                                                                                                                                                |
| „ „ „ of $F_2$ plants in the<br>late group       | <div style="display: inline-block; vertical-align: middle;"> <div style="display: inline-block; vertical-align: middle;">none shorter than 9.0 cm.</div> <div style="display: inline-block; vertical-align: middle;">majority 10.5—12. cm.</div> </div>                                                                                                                                                           |

Thus, the actual results in the present experiments suggested strongly the relation between the stem characters and flowering time to be only a physiological one.

### $F_3$ Raisings.

#### In 1912.

The sowing was done on May 7, taking 30 seeds from each individual of the following families:—

$I \times 1.4$   
 $4 \times XVI,2$

Crosses with the early flowering pure line of **G. P.**

$$\left. \begin{array}{l} \text{VIII} \times 12.1 \\ \text{VIII} \times 12.4 \\ 12 \times \text{VIII}.1 \\ 12 \times \text{VIII}.2 \end{array} \right\} \text{Crosses with the late flowering pure line of } \mathbf{G. P.}$$

Sprouting began on the 17th and ended on the 19th of the month. There were a few which sprouted after the 20th, but the flowering dates of such delayed individuals were not recorded. As already mentioned, from this year the experiments were carried on in the nursery ground.

In Tables 9 and 10, Pls. XII—XIV, we show the frequency distribution of the  $F_3$  raisings of the year 1912, arranging all families according to the similarity to their variation types. The white dots indicate the white-flowered individuals and the black dots the red-flowered ones.

#### In 1913.

In 1913, the  $F_3$  offsprings of the families  $\text{XVI} \times 3.2$ ;  $\text{XVI} \times 3.3$  and  $3 \times \text{XVI}.2$  were raised. The seeds were sown on May 9, and sprouting took place during the 3 days from the 21st to the 23rd of the month.

The results of this raising are shown in Table 11, Pls. XV—XVI.

#### Chief Facts from $F_3$ Raisings.

In the  $F_3$  raising of 1910 (Table 3) where only the progenies of those  $F_2$  individuals were raised which flowered within the variation ranges of the parent varieties (page 236), we have seen that there were several types of variation of would-be constant (homozygous) and variable (heterozygous). Now in the present  $F_3$  raisings, where the progenies of almost all individuals of each of the selected  $F_2$  families were grown, we see greatly complicated variation types, and can here again distinguish two types of variation, the constant and variable.

At first, we shall treat of the constant families. In Table 9 (Pls. XII and XIII), 19 families, from No. 1 to No. 19, are what might be taken as constant. Judging from the variation modes and means of these constant families, Nos. 1, 2 & 3 seem to be the *early constant* families, having the characteristics

of **M. P.**, and No. 19, *the late constant*, equivalent to **G. P.** Other families are more or less different from those of the parent varieties, but none are transgressive, being all intermediate between the parent varieties. Among these intermediate constants, No. 4 is very near to the early constant, and Nos. 16—18 to the late constant. We shall designate such families as *pseudo-early* and *pseudo-late constants* respectively. Other remaining intermediate constants are to be classified into two groups. Nos. 5—10 are to be grouped together as the *early intermediate constant*, and Nos. 11—15 as the *late intermediate constant*. And it will be seen that the  $F_2$  parent individuals of all families of the former group belonged to the early flowering group and those of the latter group to the late flowering group. (Compare parent classes of these families and minimum frequency classes in Table 7)

In Table 10 (Pl. XIV), Nos. 1 and 2 are considered early constant and No. 3 pseudo-early constant. The 5 families, Nos. 12—16, have the variation types very similar to that of **G. P.**, only 2 of them (No. 12 and No. 13) have a few individuals which flowered a little earlier than **G. P.** and may be designated as the pseudo-late constant. All other intermediate constants are to be classed into two groups just as in the case above mentioned. The 5 families, Nos. 4—8, all descending from the early flowered  $F_2$  plants, are considered as belonging to the early intermediate constant, and the 3 families, Nos. 9—11, descending from the late flowered  $F_2$  plants, as belonging to the late intermediate group. Thus, the mode of distribution of constant families is quite similar in both Table 9 and Table 10, but in the variation types of constant families, there is some difference between them. The variation types of the early constant and the early intermediate constant families in both Tables do not differ much from each other, but the late intermediate constant and the late constant families in Table 9, where the late pure line of **G. P.** was one of the parents, take a distinctly later position than those in Table 10 where the early pure line of **G. P.** was one of the parents. This indicates very clearly the hereditary difference of the two pure lines of **G. P.** From the similarity of distribution of constant families, how-

ever, we can safely assume that the hereditary difference of the pure line character is qualitative rather than quantitative. In other words, if the heredity of flowering time may be interpreted by the Mendelian factor theory, the number of factors from both pure lines is not different, but the quality of factors is different.

In Table 11 (Pls. XV and XVI) the would-be constant families are comparatively few. There are no early constants. Only one case is met with in each of the late and the pseudo-late constants (Nos. 11 and 10 respectively). Among the intermediate constants, the 7 families from No. 1 to No. 7 appear to belong to the early intermediate group. The variation types of these families are not all similar, and the difference among them is not so wide as to divide them again into different groups. Two families, No. 8 and No. 9, have a peculiar variation type. From the position of the variation range, they may be considered as late intermediate constants, but from the type of frequency distribution they suggest rather that they are families variable within a narrow range. Their intrinsic nature must be determined, however, only by the actual raising of the  $F_4$  offsprings (see Table 15).

Next, we shall dwell on the variable families. From the general characters of the variation types we may take 79 families, from No. 20 to No. 98, in Table 9, 38 families, from No. 17 to No. 54, in Table 10 and 73 families, from No. 12 to No. 84, in Table 11, as variable families. For the sake of easy comparison, they are arranged according to the similarity of the variation types. The following may be enumerated as remarkable facts.

1. None of the families which are the progenies of those  $F_2$  individuals which belonged to the early flowering group (No. 20 to No. 44 except 32, 33, 37, 39 and 41 in Table 9; No. 17 to No. 27 in Table 10; No. 12 to No. 32 in Table 11) have variation ranges which reach the variation range of **G. P.** Some vary only within the combined range of the early constant and the early intermediate constant, and the others within the combined range of the early constant and the late intermediate constant.
2. Among the families descended from the late-flowered  $F_2$  individuals,

there are only 6 families which vary like the above mentioned families (No. 32, 33, 37, 39 and 41 in Table 9, and No. 33 in Table 11). It is quite probable that the  $F_2$  parent plants of these 6 families belonged to the early flowering group genetically, but for some cause their flowering time had been so retarded as to make them fall into the late flowering group.

3. The variation ranges of all other variable families descended from the late-flowered  $F_2$  parents reach the variation range of **G. P.** Some vary from the range of **M. P.** to that of **G. P.**, as in the case of the  $F_2$  families; some vary within a combined range of the early intermediate constant and **G. P.**, while some show such a narrow variation range as to begin from that of the late intermediate constant.
4. There are a few exceptional families descended from the late-flowered  $F_2$  individuals, whose variation ranges do not reach that of **G. P.** but only that of the pseudo-late constant families (No. 60, 62, 67, 71 and 77 in Table 9, No. 34, 35, 47, 56 and 57 in Table 11).
5. Comparing the variation types of all variable families in Table 9 with those in Table 10 and Table 11, we did not find any difference in character between them. So, again, we can assume that the hereditary difference of the pure line character is qualitative and not quantitative.

#### $F_4$ Raisings.

##### In 1913.

This year, as it needed a comparatively large area for raising the  $F_3$ , the results of which had already been shown in Table 11 (Pls. XV and XVI), and as the available space was limited, only the  $F_4$  progenies of the following  $F_3$  families in Table 10 (Pl. XIV) were grown, as preliminary raisings:—

|        |              |     |
|--------|--------------|-----|
| No. 8  | (I × 1.4.3 ) | (A) |
| No. 18 | (I × 1.4.4 ) | (B) |
| No. 45 | (I × 1.4.14) | (C) |
| No. 43 | (I × 1.4.9 ) | (D) |
| No. 50 | (I × 1.4.13) | (E) |
| No. 38 | (I × 1.4.2 ) | (F) |

The results are shown in Tables 12 and 13.

Table 12, **A**. The parent family of this raising was one of the families designated as the early intermediate constant. Now it is proved actually that this latter designation was well chosen. The variation types of all families are almost equal, in correspondence with the assumption that the  $F_2$  grand-parent might be constant, and their variation means situated nearer to that of **M. P.** rather than in the intermediate position between **M. P.** and **G. P.**

Table 12, **B**. The parent family of this raising varied within the combined range of **M. P.** and the early intermediate constants; and within the range, there was a minimum frequency class, which makes it possible to divide the individuals into two groups, the early and late flowering. Now, in the table, it will be seen that all families descended from the early flowered parents (No. 1 to No. 6) show the variation type of the constant families which are very near to that of **M. P.** (pseudo-early)<sup>1)</sup>, and among the families descended from the late flowered parents, there are two types of variation, one varying just like the early intermediate constant (from No. 21 to No. 25), the other varying within the combined range of the pseudo-early and early intermediate constants, exactly like the parent family. These facts suggest strongly that the  $F_2$  grand-parent was monohybrid, being heterozygous in the determiners of the early (pseudo) flowering time and of the early intermediate flowering time. The ratios of the early constant, the variable, and the early intermediate constant families are 6 : 14 : 5, quite close to the expected ratios 1 : 2 : 1.

Table 13, **C, D, E, F**. All four parent families of the raisings shown here varied from the range of the early intermediate constant to that of the pseudo-late constant or of the late constant, and their variation types were all similar, having vacant classes near the middle of the range, which makes it possible to divide all individuals into two groups, the earlier and the later flowering. Now, in the actual raisings of the  $F_1$  families, we see that all

---

1) One  $F_3$  individual which flowered in 47 days produced too small a number of seeds and was discarded for the  $F_4$  raising.

3

\* No. in bracket points to parent No.

Means  
and  
Ratios

Variation means of constant families, and ratios of whites to reds to 2 groups, early-flowering and late-flowering, of variable families

2

c

t

l

s

t

l

r

c

i

c

v

l

c

t

i

s

t

s

s

1

---



parent individuals, which belonged to the early flowering group, produced the early intermediate constant families; and among the later flowered parent individuals, some produced the late or pseudo-late constant families, and some produced families which have almost equal variation types to those of the parent families. Here again, we see the variation types of segregation in the progenies of a monohybrid parent, and we can safely conclude that the  $F_2$  grand-parents were monohybrids, being heterozygous in the determiners of the early intermediate flowering time and of the late or pseudo-late flowering time. By comparing the variation means of the constant families of four different  $F_4$  progenies, however, we must assume that their  $F_2$  grand-parents were more or less different in the flowering character genetically. There is no practical difference between the variation means of both the earlier and later constant families in **E** and **F**, but the variation means of both the earlier and later constant families in **D** are smaller (earlier) than those of both constants in the former two raisings, and the variation means of the earlier constant families in **C** are larger (later) than that in **D**, while the variation means of the later constants are almost equal to that in **D**.

#### In 1914.

For this year, as more space was available for the experiment than in the previous years, we selected the following 15 families raised in 1913 (Table 11, Pls. XV and XVI):—

|                                           |                                           |
|-------------------------------------------|-------------------------------------------|
| No. 13 (XVI $\times$ 3.2.13) ( <b>G</b> ) | No. 79 (XVI $\times$ 3.3.12) ( <b>O</b> ) |
| No. 20 (XVI $\times$ 3.3.24) ( <b>H</b> ) | No. 76 (3 $\times$ XVI.2.11) ( <b>P</b> ) |
| No. 22 (XVI $\times$ 3.2.30) ( <b>I</b> ) | No. 82 (3 $\times$ XVI.2.30) ( <b>Q</b> ) |
| No. 4 (XVI $\times$ 3.3.6) ( <b>J</b> )   | No. 83 (XVI $\times$ 3.2.1) ( <b>R</b> )  |
| No. 7 (3 $\times$ XVI.2.6) ( <b>K</b> )   | No. 37 (XVI $\times$ 3.2.29) ( <b>S</b> ) |
| No. 8 (XVI $\times$ 3.2.2) ( <b>L</b> )   | No. 41 (3 $\times$ XVI.2.23) ( <b>T</b> ) |
| No. 58 (3 $\times$ XVI.2.4) ( <b>M</b> )  | No. 57 (3 $\times$ XVI.2.27) ( <b>U</b> ) |
| No. 73 (XVI $\times$ 3.35) ( <b>N</b> )   |                                           |

The sowing was done on May 4. A heavy drought followed the sowing and prevented the normal, even sprouting of the seeds. The first sprouting took place on the 14th of the month and continued gradually until the 6th of June.

In consequence of very uneven sprouting, irregularity in the flowering time followed. So we discarded all individuals which sprouted after the 24th of May, and selected those which had sprouted within the 10 days from the 14th to the 23rd of May. We calculated the representative value of their flowering times, by using the formula proposed already (page 239). The results thus obtained are shown in Table 14—18.

Table 14.

**G** and **H**. The parent families of the two raisings varied between the ranges of the early constant and early intermediate constant. Now we see in this table the same variation types as in Table 12 **B**, suggesting the former to be the variation type of the segregation in the offsprings of a monohybrid parent which was heterozygous in the determiners of the early flowering time and early intermediate flowering time.

**I**. The parent family of this raising had a similar variation type to the parent families of the above 2 raisings **G** and **H**, and the general types of variation in **I** are also similar to those shown in the case of **G** and **H**. In the present case, however, there is only one later constant family whose variation means is larger than the average means of the later constant families in **G** and **H**, suggesting it rather to be a late intermediate constant. Thus we may assume that the  $F_2$  grand-parent of **I** raising was a monohybrid which is heterozygous in determiners of the early flowering time and the late intermediate flowering time. It is quite remarkable that in the variable families of **I** raising, the white-flowered individuals are distinctly numerous toward the early part of the range, while in those of **H** raising (family Nos. 6, 7, 8, 9, 12, 13, 14 and 17) the distribution of white-flowered individuals is uniform.

Table 15.

**J**. The parent family of this raising was one of the early intermediate constants, but its variation type was rather different from the other related constants. Its maximum frequency was in the class 50. In class 51 there

were no frequencies, but in classes 52 and 53 the frequencies increased. In **J** we see that the variation types of all families are not so uniform as we have seen in Table 12 **A**. The 18 families, from No. 1 to No. 18, are very similar in their variation types, but the remaining 8 families have different variation types. The variation means of the latter are greater than those of the former. From this we can assume that the  $F_2$  grand-parent plant was not constant in a strict sense, but somewhat variable in its nature.

**K**. The parent family of this raising was also one of those which were to be designated as the early intermediate constants. Its variation range was wider than that of the parent family of **J**. Its maximum frequency class was 51, one day later than in the case of the parent family of **J**. By examining Table **K**, however, we can assume that the wider range of variation in the parent family was not genetic but environmental, because the variation types of all the families in **K** are narrower in range and very similar to one another, suggesting genetical constancy of the  $F_2$  grand-parent plant. But the difference in the position of the maximum frequency class in the parent families of **K** and **J** might have genetic meaning, because the average variation means in the **K** raising is larger (later) than that of the majority of the families in **J**. From this inequality of the variation means in both raisings, we have to conclude that all  $F_3$  families, which we have designated as the early intermediate constant, are not equal genetically.

**L**. The parent family of this raising, together with the family No. 9 in Table 11 (Pl, XV), had a peculiar variation type (see page 248). They were very likely to be the families which vary within the combined range of the early intermediate and the late intermediate constant, having their maximum frequency class in the latter constant. Now, by the actual raising of  $F_4$  progenies, we see:—

1. All  $F_4$  families are more or less different in variation type, suggesting that the  $F_2$  grand-parent was not a strictly constant one,
2. All families, however, vary only within the combined range of the previous two raisings (**J** and **K**), suggesting that the occurrence of the maxi-

imum frequency class in the parent family later than in the parent families of **J** and **K** was not genetic but environmental, i. e. the flowering times of almost all individuals grown in one row were retarded by some surrounding condition, probably by the soil property of that row. (It is remarkable that the flowering times of two families, No. 68 and No. 83 in Table 11, which were grown in the two rows next to the present family, appeared also to be retarded, especially that of No. 68, whose variation range exceeded the variation range of **G. P.** in its upper end. But by raising  $F_4$  offsprings of this latter family, it was found that this retarding in flowering was not genetic, see Table 17 **R.**).

Table 16.

**M, N** and **O**. In all three raisings shown in the present table, we meet with the variation types of segregation in offsprings of a monohybrid grandparent, producing three kinds of families, the earlier constant families, the later constant families, and the families variable within the combined range of the above two constants, just as we have seen in Table 13. According to our classification of constants, the earlier constants in this table must be of the early intermediate, while the later constants must be of the pseudo-late or late constant. The variation means of the later constants in all three raisings are very similar, but those of the earlier constants are more or less different, being in **M** the earliest, in **N** the medium, and in **O** the latest. The order of difference in the variation means in the present cases corresponds with the order of the difference in the beginning of the range in the  $F_3$  parent families (see Table 11). This indicates that each of the  $F_2$  grand-parent plants of the above three raisings contained a different determiner for the earlier constant character (early intermediate constant).

Table 17.

**P**. Here again we meet with the variation types of the monohybrid segregation. The parent family of this raising varied in a range earlier

than that of **O** in the previous table, but the earlier constant families of the present raising vary in a later range compared with **O**. It is probable that the parent family of **O** was retarded in its flowering time, as in the case of the parent family of **L**.

**Q** and **R**. The variation types of the present two raisings are also of the monohybrid segregation. The earlier constants among them are to be classified as the late intermediate constant, and the later constants have almost equal variation means as **G. P.** and are to be classified as the true late constant. The parent family of **R** (No. 83 in Table 11) varied in a later range compared with that of **Q** (No. 82), but in the table we can see practically no difference in variation between **Q** and **R**. So we can assume that the parent family of **R** was retarded in its flowering time, together with its neighbours (Families No. 8 and No. 68), probably by the soil property of the row.

Table 18.

**S** and **T**. The parent families of the 2 raisings varied within the combined range of **M. P.** and **G. P.**, as in the case of the  $F_2$  families. Now, by raising the  $F_4$  progenies, we obtained types of variation similar to the  $F_3$  raisings shown in Tables 9, 10 and 11. There are early constants (No. 1 in **S**, Nos. 1 and 2 in **T**), pseudo-early constants (No. 2 in **T**), early intermediate constants (Nos. 3, 4 and 5 in **T**), late intermediate constants (Nos. 3, 4, 5 and 6 in **T**), and late constants (Nos. 6 and 7 in **S**, No. 7 in **T**). The rest are all variable families. Those descended from the  $F_3$  individuals which were to be grouped as the early flowering, do not vary so widely as to reach the range of **G. P.**, while those which descended from the late flowering group vary in different grades of width of the variation range. (See pages 248 and 249) From these facts we may safely conclude that the  $F_4$  grand-parent plants of **S** and **T** were equal, or nearly so, in flowering character as the  $F_1$  hybrid plant. And it is noticeable that the late constant in **S** has larger variation means than that in **T**, while the parent family of **S** varied more widely than that of **T** in the upper end of the range.

**U.** The parent family of this raising had a variation type somewhat similar to that of the parent family of **M** in Table 16, but the variation types in the  $F_4$  progenies are quite different in these two raisings. The variation types in **U** are quite similar to those in **S** and **T**, except in the non-presence of the early constant. So we may assume that the  $F_2$  grand-parent of this raising was similar to the  $F_1$  hybrid plant in zygotic character, but in its  $F_3$  offsprings did not produce any early constant individual.

### Interpretation of the Experimental Results.

In the foregoing pages, we have recorded the results of our experimental work extending over eight years, and we now think that there has been accumulated sufficient material to enable us to propose an interpretation of the inheritance of the flowering time in peas.

In the first place, let us propose a hypothesis, and then let us apply it for the explanation of the results of our experiments, and see how far this hypothesis may work true. The hypothesis consists of two clauses.

1. The inheritance of flowering time in the varieties of peas which were used in the present experiments, is governed by two Mendelian alleromorphic pairs, each factor of which has a specific hereditary effect as follows:—
  - A** determines the proper flowering time of the late parent.
  - B** determines flowering a few days earlier than the proper time of the late parent and is hypostatic to **A**.
  - a** alleromorphic to **A**, determines the proper flowering time of the early parent and is epistatic to **b**.
  - b** alleromorphic to **B**, determines the flowering a few days later than the proper flowering time of the early parent.
2. Gametic contamination is caused by hybridization.

### Two-factor Hypothesis and Experimental Results.

According to the proposed two-factor hypothesis,  $F_2$  families must con-

TABLE 14—Frequency distribution in  $F_4$  (1914)

| Designation                    | Parent Class | Class Centers |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           | Means and Ratios |       |
|--------------------------------|--------------|---------------|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|-----------|------------------|-------|
|                                |              | 31            | 32 | 33 | 34 | 35 | 36 | 37 | 38 | 39 | 40 | 41 | 42 | 43 | 44 | 45 | 46 | 47 | 48 | 49 | 50 | 51 | 52 | 53 | 54 | 55 | 56 | 57 | 58 | 59 | 60 | 61 | 62 | 63 | 64        |                  | 65    |
| M P.                           |              |               | 1  | 2  | 11 | 7  | 9  | 13 | 7  |    | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           |                  | 35.84 |
| G. P.                          |              |               |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    | 5  | 11 | 10 | 11 | 10 | 7  | 10 | 1  |    | 3  | 2  | 1         |                  | 56.72 |
| Table II, No. 13 (XVI×3.2.13). | 1 (3)        | 45            |    | 2  | 11 | 5  | 5  | 2  | 2  | 1  | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           | 35.31            |       |
|                                | 2 (4)        | 45            |    | 2  | 4  | 1  | 3  | 1  | 1  | 1  | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    | 35.80     |                  |       |
|                                | 3 (9)        | 45            |    | 3  | 1  | 5  | 4  | 8  | 1  | 3  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    | 35.31     |                  |       |
|                                | 4 (19)       | 45            |    |    | 6  | 3  | 3  | 5  | 5  | 2  |    | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    | 35.44     |                  |       |
|                                | 5 (24)       | 45            |    |    | 4  | 5  | 4  | 7  | 5  |    | 2  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    | 35.44     |                  |       |
|                                | 6 (26)       | 45            |    |    | 1  | 4  | 5  | 5  | 7  | 3  | 1  | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    | 39.00     |                  |       |
|                                | 7 (27)       | 45            |    |    | 4  | 6  | 6  | 3  | 2  | 1  | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    | 35.00     |                  |       |
|                                | 8 (10)       | 48            |    |    |    | 3  | 9  | 6  | 6  | 2  | 2  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    | 39.04     |                  |       |
|                                | 9 (7)        | 49            |    |    |    | 1  |    |    |    | 2  | 6  | 8  | 5  | 1  |    |    | 3  |    |    | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           | av. 35.54        |       |
|                                | 10 (21)      | 49            |    |    |    | 1  |    | 1  |    | 2  | 1  | 5  | 8  | 1  | 1  | 1  | 1  |    |    | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           |                  |       |
|                                | 11 (25)      | 49            |    |    | 1  | 1  |    |    | 6  | 2  | 1  | 1  | 3  | 5  | 1  | 2  | 4  | 1  |    | 2  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           |                  |       |
|                                | 12 (28)      | 49            |    |    |    |    | 1  | 4  | 2  | 1  |    | 4  | 6  | 3  | 3  | 4  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           |                  |       |
|                                | 13 (39)      | 50            |    |    |    | 4  |    | 2  |    | 2  | 6  | 3  | 5  | 1  |    | 1  |    |    | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           |                  |       |
|                                | 14 (17)      | 50            |    |    |    |    | 1  | 1  |    | 2  | 5  | 4  | 5  | 4  | 1  | 3  |    | 2  |    | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           |                  |       |
|                                | 15 (14)      | 50            |    |    |    |    | 1  | 2  |    | 2  | 2  | 5  | 6  | 1  | 1  | 3  |    | 1  |    |    |    | 1  |    |    |    |    |    |    |    |    |    |    |    |    |           |                  |       |
|                                | 16 (1)       | 50            |    |    |    | 1  |    |    |    | 3  | 1  | 1  | 10 | 2  | 4  | 1  | 1  |    | 2  |    |    | 1  |    |    |    |    |    |    |    |    |    |    |    |    |           |                  |       |
|                                | 17 (16)      | 52            |    |    |    | 2  |    |    |    | 3  | 3  | 1  | 2  | 4  | 6  |    | 2  | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           |                  |       |
|                                | 18 (2)       | 51            |    |    |    | 2  | 2  | 2  | 2  |    | 1  | 3  | 3  | 2  | 1  | 5  | 1  | 3  |    |    |    | 1  |    |    |    |    |    |    |    |    |    |    |    |    |           |                  |       |
|                                | 19 (20)      | 50            |    |    |    |    |    |    |    |    | 1  | 8  | 10 | 4  | 1  | 1  |    | 1  |    | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           | 41.41            |       |
|                                | 20 (6)       | 51            |    |    |    |    |    |    |    |    | 2  | 4  | 5  | 7  | 2  | 2  |    | 1  |    |    | 2  |    |    |    |    |    |    |    |    |    |    |    |    |    |           | 42.04            |       |
|                                | 21 (8)       | 51            |    |    |    |    |    |    |    |    | 2  | 3  | 3  | 5  |    |    |    | 3  |    | 1  |    | 5  |    |    |    |    |    |    |    |    |    |    |    |    |           | 43.80            |       |
|                                | 22 (11)      | 51            |    |    |    |    |    |    |    |    | 2  | 5  | 4  |    | 2  | 3  | 1  | 2  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           | 42.52            |       |
|                                | 23 (18)      | 51            |    |    |    |    |    |    |    |    | 4  | 4  | 11 | 4  | 1  | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           | 40.91            |       |
|                                | 24 (22)      | 51            |    |    |    |    |    |    |    |    | 2  | 11 | 9  | 1  |    |    | 1  |    | 3  | 3  | 1  | 3  |    |    |    |    |    |    |    |    |    |    |    |    |           | 40.58            |       |
|                                | 25 (13)      | 50            |    |    |    |    |    |    |    |    |    | 4  | 5  | 1  |    | 5  | 2  | 1  | 3  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           | 43.89            |       |
|                                | 26 (12)      | 50            |    |    |    |    |    |    |    |    |    | 1  | 4  |    | 4  | 1  | 2  | 3  | 1  | 1  | 2  |    |    |    |    |    |    |    |    |    |    |    |    |    |           | 41.26            |       |
|                                | 27 (5)       | 50            |    |    |    |    |    |    |    |    |    | 1  | 9  | 4  | 1  | 5  |    | 3  |    | 1  | 1  | 2  |    |    |    |    |    |    |    |    |    |    |    |    |           | 42.75            |       |
|                                | 28 (20)      | 52            |    |    |    |    |    |    |    |    |    |    | 1  | 3  | 4  | 4  | 3  | 7  | 1  | 1  | 2  | 3  |    |    |    |    |    |    |    |    |    |    |    |    |           | 45.34            |       |
|                                |              |               |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           | av. 43.68        |       |
| Table II, No. 20 (XVI×3.3.24). | 1 (5)        | 45            |    |    | 2  | 8  | 6  | 14 | 2  | 3  |    | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           | 35.32            |       |
|                                | 2 (8)        | 45            |    |    | 3  | 1  | 12 | 3  | 2  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           | 39.35            |       |
|                                | 3 (26)       | 45            |    |    | 2  |    | 3  | 2  |    | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           | 35.24            |       |
|                                | 4 (29)       | 45            |    |    |    | 7  | 4  | 5  | 3  |    | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           | 35.06            |       |
|                                | 5 (14)       | 47            |    |    |    | 1  | 1  | 5  | 3  | 5  | 3  | 2  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    | 37.32     |                  |       |
|                                | 6 (1)        | 49            |    |    |    | 1  |    | 3  | 2  | 2  | 6  | 2  |    | 1  | 1  | 4  | 1  | 1  | 3  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    | av. 39.05 |                  |       |
|                                | 7 (6)        | 49            |    |    |    |    | 1  | 1  | 1  | 4  | 2  | 7  | 3  | 1  | 2  | 1  | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           |                  |       |
|                                | 8 (10)       | 49            |    |    |    | 1  |    |    |    | 1  | 2  | 1  | 7  | 1  | 2  | 1  | 1  |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |           |                  |       |





TABLE 15.—*Frequency distribution in  $F_4$  (1914)*

[illegible]

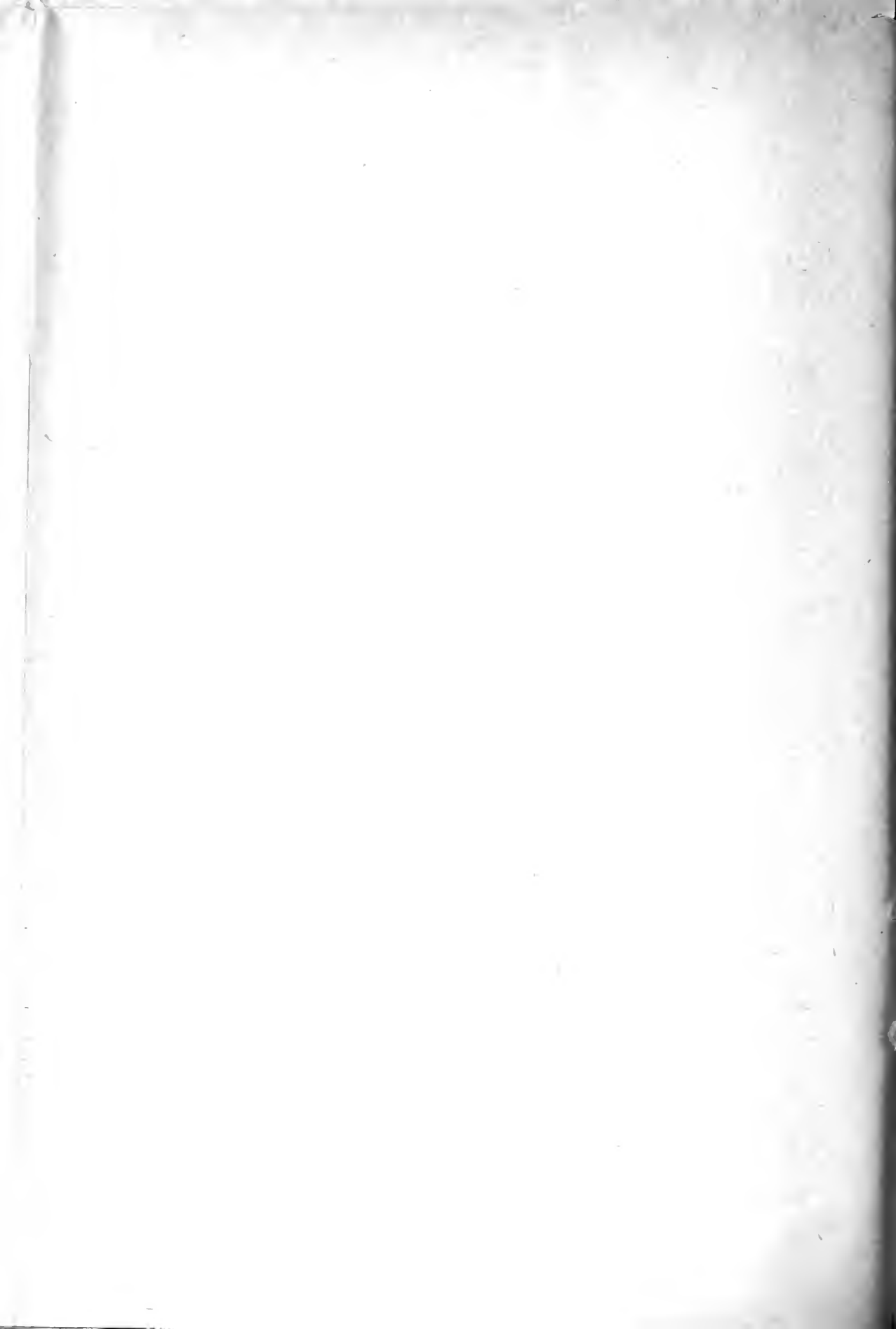


TABLE 16—Frequency distribution in  $F_4$  (1914)

[illegible]

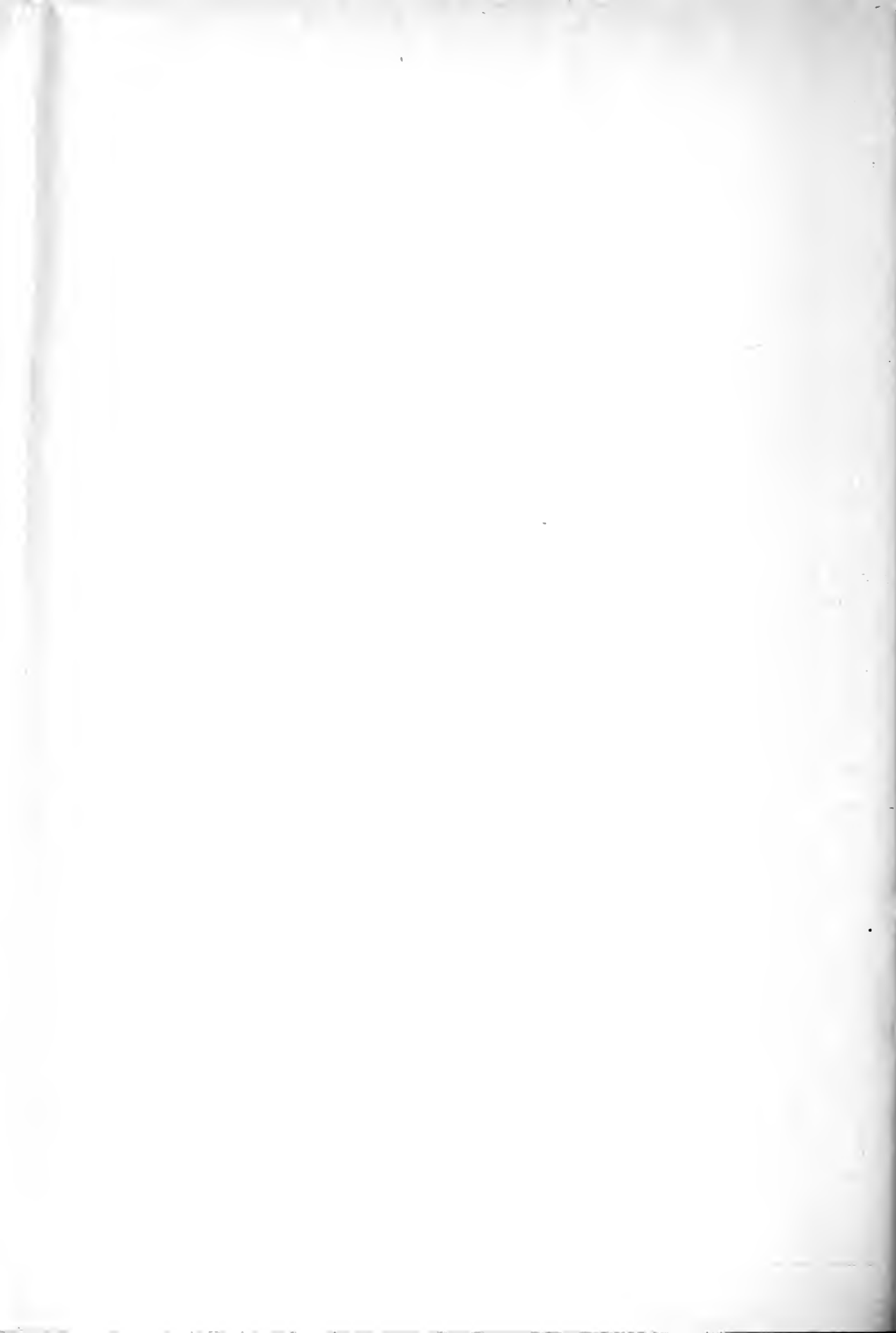


TABLE 17.—*Frequency distribution in  $F_3$  (1914)*

[illegible]



TABLE 18.—*Frequency distribution in  $F_2$  (1914)*

[illegible]



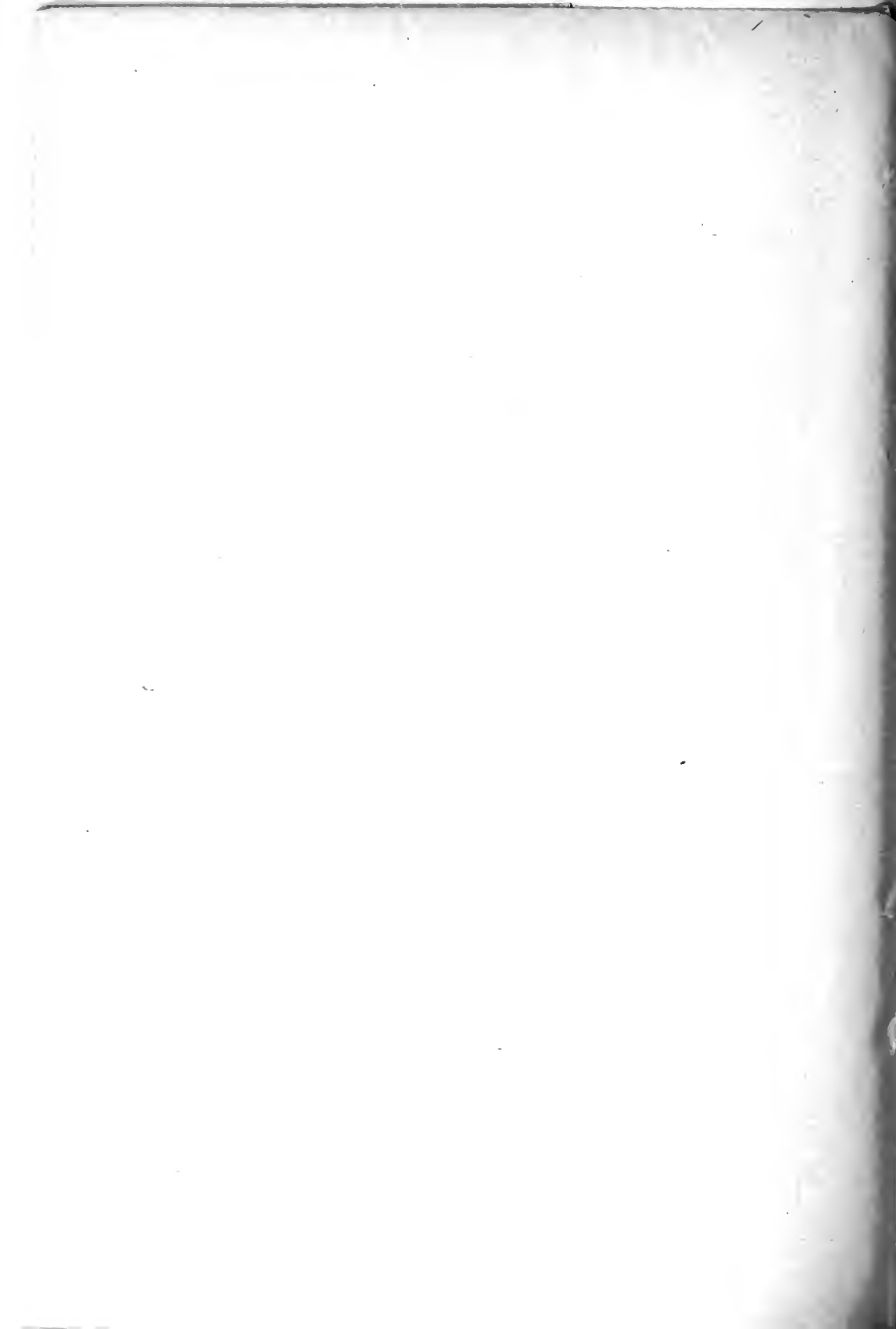


tain the following 9 different zygotic series, 5 heterozygous and 4 homozygous.

| Homozygous    |                         | Heterozygous  |                         |
|---------------|-------------------------|---------------|-------------------------|
| 1 <b>aabb</b> | } Early flowering group | 2 <b>aabB</b> | } Early flowering group |
| 1 <b>aaBB</b> |                         | 2 <b>aAbb</b> |                         |
| 1 <b>AAbb</b> | } Late flowering group  | 4 <b>aAbB</b> | } Late flowering group  |
| 1 <b>AABB</b> |                         | 2 <b>aABB</b> |                         |
|               |                         | 2 <b>AAbB</b> |                         |

Among the 5 heterozygous series, **aAbB** has the same zygotic constituent as  $F_1$ , and its flowering time must incline towards that of the late parent rather than intermediate between those of both parents (page 233 and 242); **aABB** and **AAbb**, which have become partly homozygous on account of the introduction of a factor from the late parent, must flower later than **aAbB**; **aabb** and **aAbb**, which have become partly homozygous by the introduction of one factor from the early parent, must flower earlier than **aAbB**. Two homozygous series, **aabb** and **AABB**, have the same zygotic constitution as the early and the late parent respectively. As we have seen in  $F_1$  that the dominance of lateness was incomplete, the following assumption on the flowering time of the remaining two homozygous series, **aaBB** and **AAbb**, is quite probable. **aaBB**, which is homozygous in the earlier determiners from both parents, may flower earlier, inclining towards the early parent, while **AAbb**, which is homozygous in the later determiners from both parents, may flower later, inclining towards the late parent.

In actual raisings of  $F_2$  families, we could always divide the individuals into two groups, the early flowering and the late flowering, by taking the minimum frequency class, which was at a nearly intermediate position between the parent varieties, as a line of demarkation. Now in the proposed hypothesis, we assume that the early flowering group consists of four series, **aabb**, **aaBB**, **aabB** and **aAbb**, and the late flowering group, of the remaining five series, **aAbB**, **aABB**, **AAbB**, **AAbb** and **AABB**. The ratio of the number of individuals in the former four series to those in the latter five series



tain the following 9 different zygotic series, 5 heterozygous and 4 homozygous.

| Homozygous    |                         | Heterozygous  |                         |
|---------------|-------------------------|---------------|-------------------------|
| 1 <b>aabb</b> | } Early flowering group | 2 <b>aabB</b> | } Early flowering group |
| 1 <b>aaBB</b> |                         | 2 <b>aAbb</b> |                         |
| 1 <b>AAbb</b> | } Late flowering group  | 4 <b>aAbB</b> | } Late flowering group  |
| 1 <b>AABB</b> |                         | 2 <b>aABB</b> |                         |
|               |                         | 2 <b>AAbB</b> |                         |

Among the 5 heterozygous series, **aAbB** has the same zygotic constituent as  $F_1$ , and its flowering time must incline towards that of the late parent rather than intermediate between those of both parents (page 233 and 242); **aABB** and **AAbB**, which have become partly homozygous on account of the introduction of a factor from the late parent, must flower later than **aAbB**; **aabb** and **aAbb**, which have become partly homozygous by the introduction of one factor from the early parent, must flower earlier than **aAbB**. Two homozygous series, **aabb** and **AABB**, have the same zygotic constitution as the early and the late parent respectively. As we have seen in  $F_1$  that the dominance of lateness was incomplete, the following assumption on the flowering time of the remaining two homozygous series, **aaBB** and **AAbb**, is quite probable. **aaBB**, which is homozygous in the earlier determiners from both parents, may flower earlier, inclining towards the early parent, while **AAbb**, which is homozygous in the later determiners from both parents, may flower later, inclining towards the late parent.

In actual raisings of  $F_2$  families, we could always divide the individuals into two groups, the early flowering and the late flowering, by taking the minimum frequency class, which was at a nearly intermediate position between the parent varieties, as a line of demarkation. Now in the proposed hypothesis, we assume that the early flowering group consists of four series, **aabb**, **aaBB**, **aabB** and **aAbb**, and the late flowering group, of the remaining five series, **aAbB**, **aABB**, **AAbB**, **AAbb** and **AABB**. The ratio of the number of individuals in the former four series to those in the latter five series

must be 6 : 10. The ratio of the actual numbers in the early flowering group to those in the late flowering group is as follows :—

|                        | Early group |   | Late group |
|------------------------|-------------|---|------------|
| Table 2                | 112         | : | 259        |
| Table 8 A <sub>1</sub> | 25          | : | 72         |
| Table 8 A <sub>2</sub> | 26          | : | 43         |
| Table 8 B <sub>1</sub> | 13          | : | 29         |
| Table 8 B <sub>2</sub> | 32          | : | 64         |
| Table 8 C              | 50          | : | 90         |
| <hr/>                  |             |   |            |
| Sum                    | 258         | : | 557        |
| Ratio                  | 6           | : | 12.9       |

The actual ratio exceeded the expected one, but this discrepancy is not so great as to be inadmissible. It is quite probable that such a discrepancy should occur, because, in the actual condition of plant raising, the hastening in flowering is rather rare, but its retardation is of very common occurrence, caused by environmental influences, both physiological and mechanical.

According to the present hypothesis, there must be, in the F<sub>3</sub> raisings, four kinds of constant families, **aabb**, **aaBB**, **AAbb**, and **AABB**. In Tables 9, 10 and 11 (Pls. XII—XVI), we were able to group constant families into four types,—the early constant, the early intermediate constant, the late intermediate constant, and the late constant (see page 247), except a comparatively few families, which we have designated as the pseudo-early or pseudo-late constant, and also two abnormal families, No. 8 and No. 9 in Table 11. The early constants might correspond to **aabb**, the early intermediate constants to **aaBB**, the late intermediate constant to **AAbb**, and the late constant to **AABB**, and the pseudo-early and the pseudo-late constants might be considered as contaminated early and late constants respectively. We shall discuss the two abnormal families, No. 8 and No. 9 in Table 11 later on.

In the F<sub>3</sub> variable families descended from the heterozygous F<sub>2</sub> plants, which belong to the early flowering group, we should expect two different variation types, the types of the progeny of **aabB** and of that of **aAbb**. The

type of the progeny of **aabB** should be that of the progeny of a monohybrid, varying between the early constant (**aabb**) and the early intermediate constant (**aaBB**), while that of the progeny of **aAbb**, being also of a monohybrid type, should vary between the early constant (**aabb**) and the late intermediate constant (**AAbb**). There should not occur any individuals which flower in the same period as the late constant. In actual  $F_3$  raisings, we have seen that none of the variable families descended from the early flowered  $F_2$  plants produced such a wide variation range as to reach the range of **G. P.** (page 248). It is quite difficult to divide all variable families from the early flowered  $F_2$  plants into two distinct categories according to their variation types, as the actual variation types shown in Tables 9, 10 and 11 are too complicated and irregular. But we may say, that this difficulty in distinguishing two types of variation does not positively conflict with the proposed hypothesis, because we are concerned with experiments on such a physiological character as the flowering time, which even in homozygous progenies varies somewhat, according to environmental influences.

Among  $F_3$  variable families descended from the late flowered heterozygous  $F_2$  plants, we should expect three different types of variation, that of the progeny of **aAbB**, which varies from the early constant to the late constant, as in  $F_2$  family; that of **aABB**, which, being of a monohybrid type, varies between the early intermediate constant and the late constant; and that of **AAbbB**, which is between the late intermediate constant and the late constant. In Tables 9, 10 and 11, it is difficult to divide sharply all variable families descended from late flowered  $F_2$  plants into three types, but it is certain that there are some families which represent each of the three expected types.

The upper end of the variation ranges of all  $F_3$  variable families descended from the late flowered  $F_2$  plants reaches the range of **G. P.** except a few cases as described in (2) and (4) on pages 248 and 249. Among these exceptional families, those described in (2) we have regarded already as the progenies of  $F_2$  individuals which were in the early flowering group genetically but fell into the late group, owing to some environmental influences, and as

for those described in (4), we shall try to interpret them later on, by the contamination hypothesis.

So far as variation types are concerned, we could interpret approximately all experimental results in the  $F_3$  raisings by the proposed two-factor hypothesis, admitted that some gametic contamination did take place. Now, we shall consider the numerical ratios among  $F_3$  families assumed to be in different zygotic constitutions. As it is quite difficult to distinguish clearly between the progenies of **aabB** and **aAbb**, of **aAbB** and **aABB**, and of **aABB** and **AAbB**, we shall take the progenies of **aabB** and **aAbb** as one group (variable families descended from the early flowered  $F_2$ ), and those of **aAbB**, **aABB** and **AAbB** as another (variable families from the late flowered  $F_2$ ). The ratios between four constants and two groups of variables in Tables 9 and 10 are as follows:—

|                    | aabb <sup>1)</sup> | aaBB  | AAbb  | AABB <sup>2)</sup> | Early group<br>variables | Late group<br>variables |
|--------------------|--------------------|-------|-------|--------------------|--------------------------|-------------------------|
| Table 9            | 4                  | 6     | 5     | 4                  | 25                       | 55                      |
| Table 10           | 3                  | 5     | 3     | 5                  | 11                       | 27                      |
| Sums               | 7                  | 11    | 8     | 9                  | 36                       | 82                      |
| Actual<br>Ratios   | 0.8 :              | 1.3 : | 1.0 : | 1.1 :              | 4.5 :                    | 10.2                    |
| Expected<br>Ratios | 1 :                | 1 :   | 1 :   | 1 :                | 4 :                      | 8                       |

Here we do not see much difference between actual ratios and expected. In Table 11, however, we meet with a very singular distribution of constant families. There are only two families of the late constant, the early constant being entirely absent; while there are seven families of the early intermediate constant (No. 1—No. 7), and two families (No. 8 and 9) with peculiar variation types. To attribute to chance such a deviation of actual distribution from the expected, seems to be rather conventional, but to deny absolutely that such a deviation might occur by chance may be dogmatic. So, we believe that

1) Including "pseudo-early"

2) Including "pseudo-late"

the occurrence of such deviation does not militate against proposed two-factor hypothesis.

We shall next examine the experimental results in  $F_4$  raisings. The variation types of **C**, **D**, **E** and **F** in Table 13, and **M**, **N** and **O** in Table 16 can be interpreted if we assume that the  $F_2$  grand-parents were **aABB** in their zygotic constitution, taking gametic contamination as an admitted fact (pages 251 and 254). In the same way, we can assume **B** in Table 12 and **G** and **H** in Table 14 to be the variation types of the offsprings of **aabB** (pages 250 and 252); **I** in Table 14, of **aAbb** (page 252); **P**, **Q** and **R** in Table 17, of **AAbB** (page 254); and **S**, **T** and **U** in Table 18, of **aAbB** (page 255). **A** in Table 12 and **K** in Table 15 indicate the homozygosity of their  $F_2$  grand-parents (the early intermediate constant **aaBB**). The variation types **J** and **L** in Table 15 might be interpreted as those of the progenies of the homozygous **aaBB**, if we assume that their  $F_2$  grand-parents by gametic contamination produced such offsprings as vary within a small range.

### Gametic Contamination.

It has been affirmed by the pedigree raisings of the parent varieties from 1910 to 1913, that the character of the flowering time was quite fixed and unchangeable in the parent varieties. (Tables 4, 5, 6) From this fixedness of the flowering character it should be expected, according to the common Mendelian interpretation, that all families descended from those individuals which had the same zygotic constitution, would produce the same variation type. But our actual experimental results do not fulfil this expectation, as long as we keep the proposed two-factor hypothesis.

In the  $F_3$  raisings, there has been frequent occurrence of the pseudo-early and pseudo-late constants. Such is not admissible on the two-factor hypothesis when gametic purity is retained. We took **B** in Table 12, as variation types of the segregation of **aabB**, but the variation means of all six earlier constant families are larger than that of **M**, **P**. The variation types of four raisings, **C**, **D**, **E** and **F** in Table 13 and of three raisings, **M**, **N** and **O** in

Table 15 can be nothing but those of the segregation of **aABB**, according to the proposed hypothesis. Comparing the variation types of both earlier (would-be **aaBB**) and later (would-be **AABB**) constants of these raisings, however, we see some difference among them. From the variation types of **J**, **K** and **L** in Table 15, we can not take the  $F_2$  grand-parents of these raisings as other than those with the zygotic formula **aaBB**, but within the variation types of the above three raisings we find complicated phenomena. The variation means of the majority of families in **J** are smaller than those of all families in **K**, and the variation types in **L**, in general, remind us of those types which fluctuate between two different early intermediate constants in **J** and **K**.

Now we are in a position to decide which is reasonable, to maintain the proposed two-factor hypothesis and to accept the occurrence of gametic contamination, or to assume gametic purity and propose the other hypothesis for the interpretation. Frequent occurrence of the distinct variation types of the monohybrid segregation in  $F_4$  raisings disproved positively that the inheritance of flowering time is non-Mendelian. If the inheritance be Mendelian, more than one factor must be concerned. To propose a new interpretation other than the two-factor hypothesis, it is necessary to suppose the presence of more than two-factors. To assume the presence of more than three factors, however, conflicts with the actual results. The actual numbers of the early constant families are too great, as one would expect in case more than three factors were concerned. In Table 3, two families, Nos. 3 and 4, which had descended from the  $F_2$  plants, flowered in 45 days, had almost equal variation means and ranges as **I. P.** (early constant), and two out of the three families (1, 2 and 11) which had descended from the  $F_2$  plants, flowered in 46 days varied like **I. P.** It might be concluded that all  $F_2$  individuals which had flowered in 45 days, and some of those flowered in 46, were early constant. In Table 2, there are 8 individuals flowered in 45, and 7 flowered in 46, and it might be assumed that in the  $F_2$  raising there were present more than 10 early constant individuals among 372. Such a large number would not be



expected on the more-than-three-factor hypothesis, as, on the four-factor hypothesis, there is expected only one early constant individual among 254. In Table 9, there are three early constant and one late constant families (excluding the pseudo-early and-late) among 101 families (less than  $254/2$ ), and in Table 10, 2 early constant and 3 late constant families among 54 families (far less than  $254/4$ ). And moreover, by examining the variation types shown in Tables 9, 10 and 11, one will readily be convinced of the fact that the variation types are much too simple for those of  $F_3$  progenies whose parents had more than three alleromorphic pairs.

If we assume the presence of three factors, there are two ways of interpretation; (1) supposing that all three factors have the same hereditary effect, and (2) supposing that each of the three factors has a different hereditary effect. On the former assumption, there must be 4 kinds of constant families as shown in the following zygotic formula:—

$$\begin{array}{llll}
 \text{I} \left\{ \begin{array}{l} 1 \text{ } aabbcc \\ 1 \text{ } aabbcc \\ 1 \text{ } aabbcc \end{array} \right. & \text{II} \left\{ \begin{array}{l} 1 \text{ } AAbbcc \\ 1 \text{ } aaBBcc \\ 1 \text{ } aabbCC \end{array} \right. & \text{III} \left\{ \begin{array}{l} 1 \text{ } AABbCc \\ 1 \text{ } aaBBCC \\ 1 \text{ } AAbbCC \end{array} \right. & \text{IV} \left\{ \begin{array}{l} 1 \text{ } AABbCc \\ 1 \text{ } AABbCc \\ 1 \text{ } AABbCc \end{array} \right.
 \end{array}$$

If we assume gametic purity, even on the above hypothesis, we should expect only 4 distinct constants and can not interpret the occurrence of the pseudo-early and pseudo-late constants in the  $F_3$  raisings and of those constant families whose variation means are located in various places between the variation means of the parent varieties in  $F_4$  raisings. If gametic contamination be granted, our two-factor hypothesis explains the facts obtained by our experiments far better than the three-factor hypothesis.

If we attribute a different hereditary effect to each of the three-factors, there must be 8 different constant families in  $F_3$  raising. This conflicts with the fact that we could group the intermediate constant families into only two, the early intermediate and late intermediate, in Tables 9 and 10, especially in the former. And the variation types of the variable families in  $F_3$  and  $F_4$  raisings are too simple to be interpreted on this hypothesis. If we deduce,

from the zygotic series<sup>13</sup> in  $F_2$ , various variation types in  $F_3$  and  $F_4$  and compare these types with the actual variation types in  $F_3$  and  $F_4$  raisings, one will readily see that the two-factor hypothesis is more probable than the three-factor hypothesis.

As a natural outcome of the foregoing discussion, we are convinced that our proposed hypothesis of the presence of two alleromorphic pairs and of the occurrence of gametic contamination is the most reasonable interpretation of the inheritance of the flowering time in peas. To explain the nature and cause of contamination is difficult in such experiments as those on the inheritance of flowering time. But it is certain that the contamination in our case at least is not "inconstancy" in the sense of Castle (1912). In each of the  $F_4$  raisings which showed the variation types of monohybrid segregation, all earlier constant families had almost the same variation means and ranges and also all later constant families had uniform variation types. This indicates gametic constancy in the  $F_2$  grand-parents. The hypothesis of the presence of a secondary determiner or determiners might be maintained to some extent. If we assume that the gametes from an early parent carry with primary factors (**ab**) some secondary determiner or determiners which act on the factors from a late parent as an agent or agents having the power to hasten flowering time to a slight extent, and that those from the late parent carry a secondary determiner or determiners, acting on the primary factors from the early parent as a retarding agent or agents, we can explain the occurrence of both pseudo-early and pseudo-late constant families. The variation types in **J** and **L** in Table 14 can be interpreted if we assume that their  $F_2$  grand-parents were homozygous in the primary factors (**aaBB**) but heterozygous in

| 1) | Constant | Monohybrid | Dihybrid | Trihybrid |
|----|----------|------------|----------|-----------|
| 1  | aabbcc   | 2 Aabbcc   | 4 AaBbcc | 8 AaBbCc  |
| 1  | AAbbcc   | 2 aaBbcc   | 4 aaBbCc |           |
| 1  | aaBBcc   | 2 aabbCc   | 4 AabbCc |           |
| 1  | aabbCC   | 2 AaBBcc   | 4 AaBbCC |           |
| 1  | AABbcc   | 2 AabbCC   | 4 AaBBCC |           |
| 1  | AAbbCC   | 2 AABbcc   | 4 AABbCc |           |
| 1  | aaBBCC   | 2 AAbbCc   |          |           |
| 1  | AABCC    | 2 aaBbCC   |          |           |
|    |          | 2 aaBBCC   |          |           |
|    |          | 2 AABCCc   |          |           |
|    |          | 2 AABbCC   |          |           |
|    |          | 2 AaBBCC   |          |           |

the secondary determiner or determiners. The difference of variation types between **D** and **E**, in Table 13 (see page 251), and **P** and **Q** in Table 17 (page 255) can be understood, if we assume that the  $F_2$  grand-parents of **D** and **P** become homozygous in the earlier secondary determiner or determiners, while those of **E** and **Q** become homozygous in the late secondary determiner or determiners. But the variation types of **M**, **N** and **O** in Table 15 are not to be explained by a simple assumption of the presence of a secondary determiner or determiners, as each of the above three raisings has its own proper variation types in earlier constant families, while the variation types of the later constant families are uniform in all three raisings. We hope, by further experiments, to be able to cast a clearer light on this contamination hypothesis.

#### On Transgressive Inheritance.

Throughout our experiments with peas and also with rice, we did not find a single case of transgressive inheritance. On the proposed two-factor hypothesis, however, we might expect the occurrence of such an inheritance. In the crosses between the late and early constant, or between either of these two constants and either of two intermediate constants (the early intermediate and the late intermediate), we can not expect any transgressive inheritance, but when we cross two intermediate constants (**aaBB** and **AAbb**) we should expect transgressive inheritance to take place. To state it in general terms, transgressive inheritance occurs oftener in the case of crosses between the varieties with narrower difference of character than in the case of crosses between those with wider difference. From the address of Nilsson-Ehle at the Fourth International Conference of Genetics held in Paris, 1911, we find, although he did not state it in so many words, that among his several cross breeds in wheat and oats, those breeds which were the descendants of the crosses between the varieties with comparatively wider difference in their precocity did not show transgressive variation, while those descended from the crosses between the varieties with narrower difference did. Even a cross

between the varieties with the same precocity produced very clearly the transgressive inheritance (see pp. 142-147 of *Comptes Rendus et Rapports de la Conference*). We are now conducting experiments on crosses among derivative constants, hoping in the near future to have the pleasure of publishing the results.



## II. EXPERIMENTS WITH RICE

The varieties of rice which can be grown in Sapporo, where the late frost ceases late in May and the early frost begins about the beginning of October, must be those which have extraordinary precocity. We have only a few varieties suitable for cultivation here. They are all local varieties specially bred. The difference of the ripening period among these varieties is quite insignificant. They flower and ripen at about the same time. Even the earliest varieties in southern Japan do not ripen in the vicinity of Sapporo.

In April 1907, the author procured a variety of rice, known by the name of **Kuro-Bozu**, through the kindness of Mr. S. Kato of Kinaï Substation of the Imperial Agricultural Experiment Station near Osaka. We sowed the seeds in pots in a forcing house at the beginning of May, and after one month removed the pots into a glass house. But the shooting of its ears had delayed so much as to make it impossible for us to execute crossing between it and any of the local varieties. But we succeeded in the next year, 1908, in producing their hybrids.

### Material Used and Experimental Methods.

1. Late flowering parent:— Variety name; **Kuro-Bozu** (we shall denote it as **K. P.**); a variety of *Oryza glutinosa*; awnless; tip of glumes black and with black striations on ridges of stalk.
2. Early flowering parent:— Variety name **Akagé (A. P.)**; one of the local varieties of *Oryza sativa*; with reddish brown awn; stalk without black striations.

Our experiments were carried on in a glass house. We used zinc pots 24 cm. in diameter and 35 cm. in height. In the three years,—1908, 1909 and 1910, the pots were filled with the soil from a paddy field, but from 1911 on, we filled them with fine river sand, adding a suitable quantity of artificial fertilizers. At first, 5 plants per pot were grown, but afterwards, 10 plants per pot. The space in the glass house available for the present experiments

was limited, and could accommodate 50 pots only; therefore we could not raise more than 500 individual plants each year.

Because it is difficult to record the time of flowering in the rice plant, we took a record of the time when the first spikelet appeared above the ligule of the sheath (shooting time) as the representative period of precocity. Mr. S. Kato, who has had much experience in the pollination of rice, informed the author that the rice plant is not absolutely self-pollinizing and that natural crossing often occurs. So we took care to envelope with paper bags all ears from which we were intending to take seeds for further raisings.

### F<sub>1</sub> Raisings.

In 1909

As the result of the crosses between **K. P.** and **A. P.** in the previous year, we got 9 grains in ♀ **K. P.** × ♂ **A. P.** and none in ♀ **A. P.** × ♂ **K. P.** On the 7th of May, we soaked the nine hybrid seeds with some of both parents in a forcing house. When the plantlets began to grow, we planted them in pots and put them in the glass house.

The number of days from soaking to shooting was as follows:—

| <b>K. P. × A. P.</b> |             | <b>K. P.</b>                  | <b>A. P.</b> |
|----------------------|-------------|-------------------------------|--------------|
| Pot I                | No. 1...92  | Pot III                       | No. 1...86   |
|                      | No. 2...93  |                               | No. 2...80   |
|                      | No. 3...94  |                               | No. 3...82   |
|                      | No. 4...94  |                               | No. 4...85   |
|                      | No. 5...93  |                               | No. 5...85   |
| Pot II               | No. 1...89  | Av. ....113.2                 |              |
|                      | No. 2...99  | Av. ....83.8                  |              |
|                      | No. 3...95  |                               |              |
|                      | No. 4...100 |                               |              |
| Av. ....94           |             | Average of the parents = 98.5 |              |

From this result, we see that the shooting time of the hybrid F<sub>1</sub> is intermediate between those of both parents, but inclining rather towards that of the early parent. This inclination is in the opposite direction in the case of F<sub>1</sub> in pea hybrids (pages 233 and 242).

## In 1910.

In the previous year, besides the raising of  $F_1$ , we made crosses again and got 6 hybrid grains in ♀ **K. P.** × ♂ **A. P.** and 7 grains in ♀ **A. P.** × ♂ **K. P.** We sowed these grains on May 18 directly in pots. There was one grain which failed to sprout in each of the reciprocal crosses, but we had 11  $F_1$  plants which came to maturity. The number of days from sowing to shooting of the  $F_1$  plants and of the offsprings of the parents was as follows:—

| Hybrid F <sub>1</sub> |                      |              | Parents                  |              |        |               |              |         |
|-----------------------|----------------------|--------------|--------------------------|--------------|--------|---------------|--------------|---------|
| Pot<br>No. 49         | <b>A. P. × K. P.</b> | 1...111 days | Pot<br>No. 47            | <b>A. P.</b> | 1...90 | Pot<br>No. 48 | <b>K. P.</b> | 1...122 |
|                       | "                    | 2...105      |                          | "            | 2...90 |               | "            | 2...119 |
|                       | "                    | 3...104      |                          | "            | 3...91 |               | "            | 3...122 |
|                       | "                    | 5...102      |                          | "            | 4...90 |               | "            | 4...121 |
|                       | "                    | 6...98       |                          | "            | 5...88 |               | "            | 5...126 |
| Pot<br>No. 50         | "                    | 7...102      | Av.....                  | "            | 6...88 | Av.....       | "            | 6...126 |
|                       | <b>K. P. × A. P.</b> | 1...98       |                          | "            | 7...88 |               | "            | 7...122 |
|                       | "                    | 2...103      |                          | 89.2         |        |               | 122.5        |         |
|                       | "                    | 3...103      |                          |              |        |               |              |         |
|                       | "                    | 4...102      |                          |              |        |               |              |         |
|                       |                      | 6...103      | Av. of parents.....105.8 |              |        |               |              |         |
| Av.                   |                      | 102.8        |                          |              |        |               |              |         |

From the above results, we see that there is no difference between reciprocal crosses, and that the shooting time in  $F_1$  is intermediate, inclining toward that of the early parent.

## On the Inheritance of Morphological Characters.

1. Awn: Awned dominated awnless in  $F_1$ . The dominance of the awned character over the awnless in the rice hybrids is widely known to the circle of Japanese experimenters, but, as far as we know, this seems not yet to have been published in any scientific paper. It is interesting to notice that in wheat the awnless character is dominant, but in rice the opposite is true.

2. Colour: Black colour of **K. P.** dominated  $F_1$  and all  $F_1$  plants had black awns and black striations on stalk.

3. Endosperm character: The distinction between *Oryza sativa* and *O. glutinosa* is based only on the difference of their endosperm character. The endosperm of the former contains common starch, while that of the latter contains a kind of dextrin. By husking, we can easily distinguish two kinds of endosperm at a glance. Husked grains of *O. sativa* present a light brownish and glassy appearance, and those of *O. glutinosa* a whitish and opaque appearance. We took one ear from every  $F_1$  plant, husked all grains and examined their endosperm characters. In every ear there were two kinds of grains and the ratio of *sativa* grains to *glutinosa* grains was approximately 3 : 1. These facts show that in the endosperm of rice there appears *xenia* as in the case of maize. The starch character dominates the dextrin character and these characters segregate according to the simple Mendelian law.

At this point, the author wishes to take the liberty of claiming *his priority in investigation on the existence of the immediate effect of pollen on the rice endosperm*. Having been interested in the fact that the rational interpretation of the occurrence of *xenia* in grains of some cereal crosses, such as in maize and rye, had become possible by the discovery of double fecundation by Nawaschin (1898) and Guignard (1899), the author made two crossings of rice, ♀ *glutinosa* × ♂ *sativa*, one in a green house of the College Botanic Garden in May 1901, and another in a paddy field in August of the same year. In the former crossing, he got 6 hybrid grains and in the latter, 12 hybrid grains. As these crossings were executed by cutting off the upper part of the glumes, and as the hybrid grains were exposed to the air, their color had become so darkened that it was impossible to distinguish by outward appearance whether their endosperms were starchy or of dextrin. So the hybrid grains were cut and soaked in iodine solution. All hybrid grains turned violet just as in the case of the grains of *sativa*, while self-fertilized *glutinosa* grains changed to brown color. It showed perfectly the presence of *xenia* in the rice endosperm, and the author published this result in a Japanese journal, the Journal of the Sapporo Agricultural Society, Vol. III, pp. 90-92, April, 1902. As the author left Sapporo for America and Europe



the following year, further experiments on this interesting subject were not carried out. In 1905, Moquette's paper appeared and confirmed the author's observations.

### **F<sub>2</sub> Raising.**

In 1910, the pressure of other work prevented the sowing of the seeds from the **F<sub>1</sub>** plants of the previous year at the same time with the cross grains on May 18, and we were obliged to sow them 10 days later. Adopting the method which is followed by practical farmers when the sowing has been delayed, we sowed all the grains after husking them. Germination was not bad, but many of the young plantlets were injured by mould and some were killed. So the **F<sub>2</sub>** raising of 1910 did not give any reliable results.

In 1911, we took 43 pots filled with river sand and sowed 20 seeds in each pot. After the young plants had grown 2 inches, half of them were thinned out. During the experiments, 5 pots, Nos. 4, 28, 30, 34 and 36, began to leak and could not be mended. So those plants which were grown in these pots were discarded. When **K. P.** began to shoot, a lowering of the temperature evidently affected the vegetative force of the plant. Therefore the variation range of the shooting time in **K. P.** became far wider than that in **A. P.**

Table 19 shows that the **F<sub>2</sub>** offsprings varied within the combined range of **A. P.** and **K. P.**, and there was no transgressive variation. By summing up the number of individuals in each class, we get the minimum frequency class (104), midway between the variation ranges of **A. P.** and **K. P.**, and can divide all individuals into two groups, the early-shooting and late-shooting, as we divided the early-flowering and late-flowering in the **F<sub>2</sub>** of pea hybrids. But in the present case, where the shooting period of **F<sub>1</sub>** inclined toward that of the early parent, there were a greater number of individuals in the early group than in the late group (220 : 120); while in the case of peas, where the flowering period of **F<sub>1</sub>** inclined toward the late parent, the number of individuals was greater in the late group than in the early group. (Tables 2

and 8)

Tschermak records in Fruwirth's "Züchtung der landwirtschaftlichen Kulturpflanzen" Vol. 4, (2 Auf.) pp. 176, 238 and 314, that in wheat, barley and rye, the early ripening character seems to be partially dominant, and in Table 39 of Emerson and East's work (1912) on maize, we see that there is a minimum frequency class within the variation ranges of the two  $F_2$  progenies (1127) and (1128), and the number of individuals in the early flowering group is greater than that in the late flowering group. From these facts, we venture to propose that, in cereals, incomplete dominance of the early ripening may be a law. We did not forget to observe the relation between morphological characters and shooting time in the  $F_2$ , but could not find any correlation between them, as we had seen in the case of peas between the flowering time and the colour of their flowers.

### $F_3$ Raising.

From 80 offsprings of **K. P.**  $\times$  **A. P.** 1 (Table 19), we selected 46 individuals with different shooting dates, and raised 10  $F_3$  offsprings of each individual, together with 20 individuals of each parent variety. In this year, the lowering of the temperature during the shooting time of **K. P.** was greater than in the previous year. Consequently, the shooting of **K. P.** individuals occurred slowly, and one of them and three of hybrid  $F_3$  individuals (in families 35, 45 and 46) did not eventually shoot.

The frequency distributions of the number of days from sowing to shooting is shown in Table 20. From this table, we can not draw any definite conclusion, except that there occurred four families, Nos. 1, 2, 45 and 46, which produced variation types quite similar to that of either of the parent varieties, suggesting that their  $F_2$  parents had returned, in shooting character, to the original parents.

### $F_4$ Raising.

From the result of the  $F_3$  raising in the previous year, we were convinced

TABLE 19.—Frequency distribution of number of days from sowing to shooting in  $F_2$  of crosses between K. P. and A. P. (Rice)

[illegible]

TABLE 20.—Frequency distribution in  $F_3$  (Progenies of  $K. P. \times A. P. 1.$ )

\* Antecedent No. of pot; subsequent, No. of plant in the pot.

× Every pot signed with this mark contained one individual which did not come to shoot.

TABLE 21.—Frequency distribution in  $F_3$ .[illegible]

\* 4 Individuals did not come to shoot.



that it would be rather impossible to expect any definite interpretation of the inheritance of shooting character in rice, by raising such a small number of  $F_4$  individuals as 500. But hoping that we might reach some interesting conclusion by comparing the results obtained in rice with those in peas, we took four  $F_3$  families, Nos. 4, 5, 10 and 16 (Table 20), which showed some similarity in variation types and selected 23 individuals from them, taking care to select those which were different in shooting dates. Twenty offsprings of each selected plant were raised.

As the shooting of **K. P.** was too late in the two previous years, the sowing of the seeds was done in wooden boxes placed in a forcing house on April 20. After the temperature in the glass house had become high enough for rice, the plantlets were transplanted in pots. In spite of this precaution, the plants did not shoot early, because of the unusually low temperature which prevailed during the summer, and which caused an almost total failure in the paddy culture in the vicinity of Sapporo. In our raising, **A. P.** came to shoot a little later than in the previous two years, and **K. P.** did not shoot at all, even in the glass house. Five individuals of the family No. 5 of **C** Table 21, also failed to shoot.

Table 21 shows the results of this  $F_4$  raising.  $F_2$  grand-parents of two raisings of **A** and **B** appeared on the same day and their  $F_3$  parent families varied in a similar type, but in the present table their variation types are not similar. Those of **B** suggest strongly monohybrid segregation, family No. 2 being an earlier constant, families Nos. 3 and 7 later constants, and the three families, Nos. 5, 1 and 10, variable between two constants. In **A**, the variation types are quite different. There is one family which seems to be a constant (No. 6). And the variation types of the remaining variable families are not those which would be expected in monohybrid segregation. In both raisings of **C** and **D**, we see also variation types of non-monohybrid segregation.

### Interpretation.

By comparing the experimental results in rice with those in peas, we see

a parallelism between them to some extent. The flowering time (shooting time in rice) of the first generation of a hybrid was not just intermediate between parent varieties. It inclined towards one of the parents, in peas towards the late parent and in rice towards the early parent. In  $F_2$ , the variation range extended within the combined ranges of both parents, having the minimum frequency class in the middle. When all  $F_2$  individuals were arranged into two groups, the early flowering and the late flowering, using the minimum frequency class as a demarkation line, the group towards which  $F_1$  inclined contained a greater number of individuals than the other group,—in the rice the early group and in peas the late group. In  $F_3$  raisings of rice hybrids, there appeared few families which seemed to be constant, though the distribution of these apparently constant families was not similar to that in the peas, and in  $F_4$  raisings we had one case of variation types closely suggesting those of monohybrid segregation (**B** Table 21).

From the above cited facts, we may safely assume that the inheritance of the shooting character in rice is not different from that of the flowering character in peas in its underlying principle and is interpretable by the multiple-factor hypothesis. But the two-factor hypothesis which was proposed for the interpretation of the inheritance of the flowering time in peas is not applicable in the case of rice, because of the following two facts.

1. The actual number of  $F_2$  individuals which seem to be in the same zygotic constitution as the original parents is much smaller than would be expected on the two-factor hypothesis. From actual results in  $F_3$  raising, we might assume those  $F_2$  individuals which shot forth within the variation range of the late parent as the late constant, and those which shot forth during the two days, 88 and 89, as the early constant. The actual number of the former is 4 and that of the latter 5, while the expected number of each of the early and late constant on the two-factor hypothesis is  $340/16 = 21.2$ .

2. According to the two-factor hypothesis, all variable families in  $F_3$  have to be divided into two categories, monohybrid segregates (descendants of **aabB**, **aAbb**, **aABB** or **AAbB**) and dihybrid segregates (descendants of

**aAbB**, same as  $F_1$ ). From the results of the raising of  $F_4$  offsprings of those  $F_3$  families such as No. 4 and No 5 of Table 20 which could be assumed, from their parent classes and their variation types, not to be descendants of  $F_2$  having the same zygotic constitution as  $F_1$  (**aAbB**), we should expect variation types of monohybrid segregation only. In the actual  $F_4$  raisings, however, this expectation was not fulfilled. In Table 21, the variation types in **B** suggest those of monohybrid segregation, but those in **A** are too complicated to be regarded as monohybrid segregation.

Then, by assuming the three-factor hypothesis, can these facts be explained? Although our experiments were conducted on too small a scale to supply enough material for a discussion of this subject, so far as our experiments go, the three-factor hypothesis does not conflict with actual results. As we have noted above, among 340  $F_2$  individuals (Table 19), 5 which shot forth during the two days 88 and 89, might be taken as the early constants and 4 which shot forth within the variation range of the late parent, as the late constants. These numbers of both early and late constant  $F_2$  individuals are in admirable correspondence with the calculated number 5.3 (340/64), according to the three-factor hypothesis. The variation types of **A** and **B** in Table 21, which were not conceivable on the two-factor hypothesis, are quite acceptable on the three-factor hypothesis, when we take  $F_2$  grand-parent of **B** as monohybrid such as **aabBcc**, **aabbC**, or **aAbbcc**, (see foot notes on page 264), and that of **A**, as a dihybrid such as **aabBcC**, **aAbbcC** or **aAbBcc**.

Here we wish to propose the three-factor hypothesis for the interpretation of the inheritance of the shooting character in the rice varieties which we have used in our present work.

Owing to climatic conditions in Sapporo, experiments on a sufficiently large scale were not possible by the author, but he hopes at some future date, with the co-operation of some investigator in southern Japan, to obtain more conclusive results.



### III. ON THE GAMETIC COUPLING BETWEEN FLOWERING TIME AND FLOWER COLOUR IN PEAS.

Lock (1904 and 1905) noticed in his experiments with pea hybrids the correlation between white flowers and early flowering, red flowers and late flowering. Afterwards Tschermak (1910) saw the same correlation in his experiments. But neither tried to explain this correlation genetically. Throughout the present experiments we also have noticed this correlation. In all  $F_2$  families, there was excess of white-flowered to red-flowered individuals in the early flowering group, but in the late flowering group, on the contrary, a very small number of white flowers was present (Tables 2 and 8). In variable families of  $F_3$  and  $F_4$  raisings, the same phenomenon prevailed, except in a very few cases. We shall consider now whether this correlation may be interpreted on the hypothesis of gametic coupling or not.

For the interpretation of the inheritance of the flowering time in peas, we proposed two alleromorphic pairs, **a, A** and **b, B**. If we assume that one character is determined by two Mendelian factors, our method of treatment must be different from that in the cases of gametic coupling hitherto investigated. We must consider at first, whether the flower colour couples exclusively with either one of the two factors, or independently with each of them, or whether the coupling takes place when two-factors are present together in the gametes. Studying carefully the distribution of different coloured individuals throughout the experimental results, we are quite convinced, that the latter two assumptions must be abandoned. By assuming that the factor for red colour couples with the more powerful factor **A** from the late parent, all the observed results can be adequately explained. We shall show in the following paragraphs the possibility of this interpretation.

If we assume gametic coupling between the factor for red colour and the factor **A** for flowering time on the 7 : 1 basis, we should expect a distribution of whites and reds in the two flowering groups of  $F_2$  families as follows:—



| Early flowering group               |            | Late flowering group                |            |
|-------------------------------------|------------|-------------------------------------|------------|
|                                     | W. R.      |                                     | W. R.      |
| 1 <b>abab</b> = $7+1 \times 7+1$    | = 49 : 15  | 1 <b>ABAB</b> = $1+7 \times 1+7$    | = 1 : 63   |
| 2 <b>abaB</b> = $2(7+1 \times 7+1)$ | = 98 : 30  | 2 <b>ABAb</b> = $2(1+7 \times 1+7)$ | = 2 : 126  |
| 2 <b>abAb</b> = $2(7+1 \times 1+7)$ | = 14 : 114 | 2 <b>ABaB</b> = $2(1+7 \times 7+1)$ | = 14 : 114 |
| 1 <b>aBaB</b> = $7+1 \times 7+1$    | = 49 : 15  | 1 <b>AbAb</b> = $1+7 \times 1+7$    | = 1 : 63   |
|                                     |            | 2 <b>ABab</b> = $2(1+7 \times 7+1)$ | = 14 : 114 |
|                                     |            | 2 <b>AbaB</b> = $2(1+7 \times 7+1)$ | = 14 : 114 |
| Sums                                |            | 210 : 174                           | 46 : 594   |
| Ratios                              |            | 4.6 : 3.8                           | 1 : 12.9   |

Counting the numbers of whites and reds in actual  $F_2$  raisings, we get the following results:—

|                                   | Early flowering group |     | Late flowering group |      |
|-----------------------------------|-----------------------|-----|----------------------|------|
|                                   | white                 | red | white                | red  |
| Table 2 .....                     | 64                    | 48  | 12                   | 247  |
| Table 8 <b>A</b> <sub>1</sub> ... | 14                    | 11  | 7                    | 65   |
| <b>A</b> <sub>2</sub> ...         | 19                    | 7   | 3                    | 40   |
| <b>B</b> <sub>1</sub> ...         | 10                    | 3   | 0                    | 29   |
| <b>B</b> <sub>2</sub> ...         | 24                    | 8   | 5                    | 59   |
| <b>C</b> .....                    | 26                    | 24  | 8                    | 82   |
| Sums                              | 157                   | 101 | 35                   | 522  |
| Ratios                            | 4.5                   | 2.9 | 1.0                  | 14.9 |

These observed ratios are sufficiently close to the expected ones. Next, when we take some of the  $F_3$  families (Tables 9—11, Pls. XII—XVI), which vary from the early constant to the late constant and which are assumed to be the progenies of  $F_2$  plants, whose zygotic constitution might be **aAbB**, and when we examine the distribution of different coloured individuals in each of them, we get the following results:—

|          |           | Early flowering group |          | Late flowering group |          |
|----------|-----------|-----------------------|----------|----------------------|----------|
|          |           | white                 | red      | white                | red      |
| Table 9  | Family 45 | ... 6                 | ..... 2  | ..... 3              | ..... 19 |
|          | " 46      | ... 6                 | ..... 5  | ..... 5              | ..... 14 |
|          | " 47      | ... 5                 | ..... 8  | ..... 0              | ..... 13 |
|          | " 48      | ... 6                 | ..... 5  | ..... 1              | ..... 14 |
|          | " 59      | ... 7                 | ..... 7  | ..... 0              | ..... 12 |
|          | " 50      | ... 4                 | ..... 5  | ..... 0              | ..... 19 |
|          | " 51      | ... 0                 | ..... 8  | ..... 6              | ..... 13 |
|          | " 52      | ... 8                 | ..... 5  | ..... 1              | ..... 16 |
|          | " 54      | ... 4                 | ..... 6  | ..... 4              | ..... 16 |
|          | " 56      | ... 6                 | ..... 5  | ..... 0              | ..... 14 |
|          | " 57      | ... 6                 | ..... 4  | ..... 0              | ..... 17 |
|          | " 58      | ... 5                 | ..... 4  | ..... 1              | ..... 20 |
|          | " 69      | ... 7                 | ..... 7  | ..... 2              | ..... 14 |
|          | " 60      | ... 6                 | ..... 1  | ..... 0              | ..... 16 |
|          | " 61      | ... 7                 | ..... 1  | ..... 0              | ..... 18 |
| Table 10 | Family 28 | ... 8                 | ..... 4  | ..... 2              | ..... 15 |
|          | " 29      | ... 8                 | ..... 6  | ..... 3              | ..... 12 |
|          | " 30      | ... 4                 | ..... 7  | ..... 0              | ..... 18 |
|          | " 31      | ... 6                 | ..... 7  | ..... 0              | ..... 16 |
|          | " 32      | ... 7                 | ..... 4  | ..... 0              | ..... 19 |
|          | " 33      | ... 4                 | ..... 5  | ..... 1              | ..... 19 |
|          | " 34      | ... 9                 | ..... 4  | ..... 1              | ..... 14 |
|          | " 35      | ... 9                 | ..... 5  | ..... 1              | ..... 13 |
|          | " 37      | ... 1                 | ..... 10 | ..... 2              | ..... 18 |
| Table 11 | Family 34 | ... 3                 | ..... 2  | ..... 1              | ..... 23 |
|          | " 35      | ... 4                 | ..... 5  | ..... 2              | ..... 18 |
|          | " 36      | ... 3                 | ..... 5  | ..... 2              | ..... 18 |
|          | " 37      | ... 3                 | ..... 4  | ..... 5              | ..... 17 |
|          | " 38      | ... 8                 | ..... 1  | ..... 9              | ..... 12 |
|          | " 41      | ... 9                 | ..... 0  | ..... 1              | ..... 16 |
|          | " 42      | ... 5                 | ..... 3  | ..... 4              | ..... 17 |
|          | " 43      | ... 5                 | ..... 2  | ..... 1              | ..... 21 |
|          | " 44      | ... 9                 | ..... 4  | ..... 1              | ..... 14 |
|          | " 45      | ... 6                 | ..... 2  | ..... 4              | ..... 19 |
|          | " 46      | ... 1                 | ..... 4  | ..... 5              | ..... 17 |
|          | " 47      | ... 4                 | ..... 1  | ..... 3              | ..... 22 |
| Sums     |           | 199.....              | 158      | 71                   | 593      |
| Ratios   |           | 2.8                   | 2.2      | 1.0                  | 8.4      |

In the above table, we meet with so large a number of whites in the late flowering group, that, when we take it as the unit and calculate ratios, the result differs much from the expected. By considering, however, that on account of environmental influences the retarding of the flowering time is of common occurrence and that some individuals belonging to the early group genetically may very often fall thereby into the late group, we may take the number of reds in the early group as the unit and calculate ratios. Then we get the following ratios:—

|          | Early group |     | Late group |     |
|----------|-------------|-----|------------|-----|
|          | W.          | R.  | W.         | R.  |
| Observed | 1.3         | 1.0 | 0.45       | 3.8 |
| Expected | 1.2         | 1.0 | 0.26       | 3.3 |

Here, except whites in the late group, the ratios are quite close to the expected.

Thus, our proposed gametic coupling is practically confirmed, but before we draw a definite conclusion, we have to prove the following two propositions:—

1. Non-presence of the correlation in  $F_3$  and  $F_4$  variable families, which are the descendants of the  $F_2$  plants which have become homozygous in the factor **A** or **a** (**aabB** or **AAbB**).
2. The occurrence of the zygotic series 49 : 15 : 15 : 177 in  $F_3$  and  $F_4$  variable families which are the descendants of the  $F_2$  plants heterozygous only in the factor **A** (**aAbb** or **aABB**).

(1) As we have already noted on page 259, the distinction between the supposed **aabB** families and the supposed **aAbb** families in  $F_3$  raisings (Tables 9, 10 and 11) is not clear, but by careful examination of the variable families descended from the early-flowered  $F_2$  plants, we see that those families which have relatively narrow ranges of variation and which suggest the progenies of **aabB** parents, have uniform distribution of white-flowered individuals and indicate non-presence of the correlation (families, Nos. 26, 27 and 28 in Table 9; Nos. 19, 20, 21 and 22 in Table 10; No. 20 in Table 11). In Table 14 **H**, where the results of the raising of  $F_4$  progenies of the family No. 20 Table

11 are shown, we see again uniform distribution of whites in variable families (page 252). Family No. 21 in Table 11 appears to conflict with the present hypothesis, because the distribution of whites in it is quite uneven, while from its parent class and variation type we have to assume it to be progeny of **aabB**. On the other hand, we may also assume it to be the progeny of **aAbb**, deducing from the example of the similar typed family No. 22 which are shown to be the segregates of **aAbb** by raising  $F_4$  progenies (Table 14).

Among those  $F_3$  families which are to be taken as the segregates of **AAbB**, the majority are homozygous red. Only one family, No. 81 of Table 11, which is variable in colour, has uniform distribution of whites. No. 79 of Table 11 seems to be the segregates **AAbB**, but by  $F_4$  raising it was proved to be those of **aABB** (Table 16 O).

(2) According to the proposed hypothesis,  $F_2$  plants which were homozygous in the factor **B** or **b** but heterozygous in the factors **A** and **a** and also in colour, must produce such  $F_3$  families which have the following distribution of different coloured individuals.

| Early group (earlier constants) |     | Late group (variables & later constants) |     |
|---------------------------------|-----|------------------------------------------|-----|
| white                           | red | white                                    | red |
| 49                              | 15  | 15                                       | 177 |

And when we raise  $F_4$  progenies of these  $F_3$  families, there must appear the same distribution of different coloured individuals in variable families.

As the variation types of the supposed **aAbb** segregates in  $F_3$  raising in 1913 (Table 11) are irregular and do not allow us to distinguish two groups, the early flowering and the late flowering, by taking a definite minimum frequency class, we shall take only Tables 9 and 10, and examine the distribution of different coloured individuals in those families which suggest the segregates of **aAbb**.

|                           |           | Early group |     | Late group |     |
|---------------------------|-----------|-------------|-----|------------|-----|
|                           |           | white       | red | white      | red |
| Table 9<br>(Min. Fr.=48)  | Family 31 | 7           | 0   | 1          | 20  |
|                           | " 32      | 6           | 5   | 0          | 18  |
|                           | " 33      | 8           | 1   | 1          | 20  |
|                           | " 34      | 7           | 1   | 2          | 20  |
|                           | " 35      | 3           | 1   | 2          | 13  |
|                           | " 36      | 4           | 1   | 0          | 23  |
|                           | " 37      | 1           | 1   | 0          | 25  |
|                           | " 38      | 7           | 0   | 1          | 20  |
|                           | " 39      | 5           | 0   | 1          | 14  |
|                           | " 40      | 2           | 2   | 2          | 17  |
| Table 10<br>(Min. Fr.=49) | Family 24 | 7           | 2   | 1          | 18  |
|                           | " 25      | 5           | 0   | 3          | 25  |
|                           | " 26      | 5           | 2   | 2          | 22  |
| Sums                      |           | 67          | 16  | 16         | 255 |

Here we see an exactly equal number of early reds and late whites, as was expected, but the number of both early whites and late reds is greater than the expected. Then, turning to **I** Table 14 in which the variation types were assumed to be those of the descendants of **aAbb**  $F_2$  plant, let us examine the distribution of whites and reds in variable families.

|        |    | Early group |     | Late group (Min. Fr.=39) |     |
|--------|----|-------------|-----|--------------------------|-----|
|        |    | white       | red | white                    | red |
| Family | 10 | 6           | 1   | 0                        | 11  |
| "      | 11 | 7           | 2   | 4                        | 15  |
| "      | 12 | 3           | 5   | 3                        | 9   |
| "      | 13 | 2           | 0   | 1                        | 14  |
| "      | 14 | 3           | 5   | 2                        | 19  |
| "      | 15 | 6           | 2   | 1                        | 16  |
| "      | 17 | 4           | 2   | 1                        | 18  |
| "      | 18 | 6           | 1   | 3                        | 17  |
| "      | 19 | 4           | 0   | 2                        | 16  |
| "      | 20 | 8           | 1   | 2                        | 14  |
| "      | 21 | 3           | 2   | 1                        | 17  |
| "      | 23 | 2           | 0   | 1                        | 13  |
| "      | 24 | 5           | 0   | 5                        | 18  |
| Sum    |    | 59          | 21  | 26                       | 197 |

Here again we see that the numbers of early reds and late whites are quite close, but the numbers of early whites and late reds are smaller than the expected. Now let us add the above two sums, then we shall get a very close correspondence between the observed and the expected.

|          | Early |     | Late  |     |
|----------|-------|-----|-------|-----|
|          | white | red | white | red |
|          | 67    | 16  | 16    | 255 |
|          | 59    | 21  | 26    | 197 |
|          | 126   | 37  | 42    | 452 |
| Expected | 137   | 42  | 42    | 495 |

Next, let us look at the distributions of whites and reds in the families which were assumed to be the segregates of **aABB**. To do this, we shall take only those families of  $F_4$  raisings, because it is quite difficult to distinguish **aABB** families from **aAbB** families in  $F_3$  raisings.

|                                     |          | Early group |     | Late group |     |
|-------------------------------------|----------|-------------|-----|------------|-----|
|                                     |          | white       | red | white      | red |
| Table 13, <b>D</b><br>Min. Fr. = 55 | Family 4 | 8           | 2   | 2          | 15  |
|                                     | " 5      | 11          | 2   | 3          | 12  |
|                                     | " 6      | 8           | 1   | 3          | 20  |
|                                     | " 7      | 5           | 0   | 2          | 20  |
|                                     | " 8      | 4           | 1   | 2          | 21  |
|                                     | " 10     | 10          | 0   | 2          | 17  |
|                                     | " 11     | 6           | 0   | 3          | 19  |
|                                     | " 12     | 5           | 0   | 0          | 24  |
|                                     | " 13     | 5           | 2   | 0          | 21  |
|                                     | " 14     | 8           | 1   | 1          | 14  |
|                                     | " 15     | 9           | 0   | 0          | 21  |
|                                     | " 16     | 3           | 0   | 0          | 24  |
|                                     | " 17     | 5           | 0   | 1          | 24  |
|                                     | Sums     | 87          | 9   | 19         | 252 |

|                                     |          |    |    |   |     |
|-------------------------------------|----------|----|----|---|-----|
| Table 13, <b>E</b><br>Min. Fr. = 58 | Family 8 | 2  | 2  | 2 | 24  |
|                                     | " 9      | 9  | 1  | 0 | 18  |
|                                     | " 10     | 5  | 3  | 0 | 20  |
|                                     | " 11     | 5  | 2  | 1 | 20  |
|                                     | " 12     | 8  | 0  | 0 | 17  |
|                                     | " 13     | 3  | 0  | 1 | 24  |
|                                     | " 15     | 3  | 0  | 1 | 25  |
|                                     | " 16     | 6  | 1  | 0 | 23  |
|                                     | " 17     | 2  | 1  | 3 | 24  |
|                                     | " 18     | 7  | 2  | 0 | 20  |
|                                     | " 19     | 5  | 1  | 1 | 20  |
|                                     | Sums     | 55 | 14 | 9 | 235 |

|                                     |           | Early group |     | Late group |     |
|-------------------------------------|-----------|-------------|-----|------------|-----|
|                                     |           | white       | red | white      | red |
| Table 13, <b>F</b><br>Min. Fr. = 59 | Family 11 | 5           | 0   | 1          | 25  |
|                                     | " 12      | 5           | 4   | 3          | 17  |
|                                     | " 13      | 3           | 1   | 1          | 25  |
|                                     | " 14      | 3           | 3   | 0          | 21  |
|                                     | " 15      | 6           | 0   | 1          | 23  |
|                                     | " 17      | 7           | 1   | 1          | 20  |
|                                     | " 18      | 8           | 0   | 0          | 21  |
|                                     | " 19      | 9           | 1   | 0          | 18  |
|                                     | " 20      | 9           | 4   | 0          | 17  |
|                                     | " 21      | 2           | 2   | 0          | 24  |
|                                     | " 22      | 7           | 0   | 2          | 21  |
|                                     | " 23      | 3           | 0   | 2          | 25  |
|                                     | Sums      | 67          | 16  | 11         | 257 |

|                                     |           |    |   |    |     |
|-------------------------------------|-----------|----|---|----|-----|
| Table 16, <b>M</b><br>Min. Fr. = 47 | Family 10 | 5  | 0 | 2  | 9   |
|                                     | " 11      | 4  | 1 | 0  | 20  |
|                                     | " 12      | 6  | 0 | 3  | 14  |
|                                     | " 14      | 1  | 0 | 4  | 21  |
|                                     | " 15      | 5  | 1 | 6  | 16  |
|                                     | " 16      | 8  | 0 | 1  | 19  |
|                                     | " 17      | 7  | 0 | 2  | 19  |
|                                     | " 18      | 8  | 1 | 1  | 15  |
|                                     | " 19      | 3  | 0 | 3  | 16  |
|                                     | " 20      | 11 | 2 | 2  | 14  |
|                                     | " 21      | 2  | 4 | 2  | 20  |
|                                     | " 22      | 4  | 0 | 1  | 21  |
|                                     | Sums      | 69 | 9 | 28 | 216 |

|                                       |        | Early group |     | Late group |     |     |               | Early group                                                          |            | Late group |     |      |      |
|---------------------------------------|--------|-------------|-----|------------|-----|-----|---------------|----------------------------------------------------------------------|------------|------------|-----|------|------|
|                                       |        | white       | red | white      | red |     |               | white                                                                | red        | white      | red |      |      |
| Table 16, <b>N</b> .<br>Min. Fr. = 50 | Family | 10          | 4   | 3          | 1   | 18  | Total<br>Sums | <b>D</b><br><b>E</b><br><b>F</b><br><b>M</b><br><b>N</b><br><b>O</b> | 87         | 9          | 19  | 252  |      |
|                                       | "      | 11          | 4   | 2          | 3   | 18  |               |                                                                      | 55         | 14         | 9   | 235  |      |
|                                       | "      | 12          | 7   | 0          | 2   | 21  |               |                                                                      | 67         | 16         | 11  | 257  |      |
|                                       | "      | 13          | 6   | 2          | 0   | 13  |               |                                                                      | 69         | 9          | 28  | 216  |      |
|                                       | "      | 14          | 6   | 3          | 1   | 18  |               |                                                                      | 48         | 29         | 14  | 166  |      |
|                                       | "      | 16          | 4   | 0          | 2   | 21  |               |                                                                      | 61         | 27         | 40  | 235  |      |
|                                       | "      | 17          | 6   | 7          | 0   | 12  |               | Sums                                                                 | 387        | 104        | 121 | 1361 |      |
|                                       | "      | 18          | 5   | 3          | 3   | 17  |               |                                                                      | Expected { | 395        | 121 | 121  | 1428 |
|                                       | "      | 20          | 4   | 5          | 1   | 17  |               |                                                                      |            | 340        | 104 | 104  | 1227 |
|                                       | "      | 21          | 2   | 4          | 1   | 11  |               |                                                                      |            | Av. 368    | 112 | 112  | 1328 |
|                                       |        | Sums        | 48  | 29         | 14  | 166 |               |                                                                      |            |            |     |      |      |
| Table 16, <b>O</b> .<br>Min. Fr. = 49 | Family | 6           | 8   | 2          | 5   | 14  |               |                                                                      |            |            |     |      |      |
|                                       | "      | 7           | 2   | 0          | 2   | 19  |               |                                                                      |            |            |     |      |      |
|                                       | "      | 9           | 4   | 1          | 4   | 15  |               |                                                                      |            |            |     |      |      |
|                                       | "      | 10          | 3   | 3          | 3   | 13  |               |                                                                      |            |            |     |      |      |
|                                       | "      | 11          | 6   | 3          | 2   | 17  |               |                                                                      |            |            |     |      |      |
|                                       | "      | 12          | 2   | 1          | 4   | 19  |               |                                                                      |            |            |     |      |      |
|                                       | "      | 13          | 5   | 3          | 5   | 19  |               |                                                                      |            |            |     |      |      |
|                                       | "      | 16          | 11  | 1          | 1   | 18  |               |                                                                      |            |            |     |      |      |
|                                       | "      | 17          | 8   | 1          | 1   | 17  |               |                                                                      |            |            |     |      |      |
|                                       | "      | 18          | 4   | 2          | 1   | 16  |               |                                                                      |            |            |     |      |      |
|                                       | "      | 19          | 2   | 6          | 4   | 17  |               |                                                                      |            |            |     |      |      |
|                                       | "      | 20          | 2   | 3          | 2   | 17  |               |                                                                      |            |            |     |      |      |
|                                       | "      | 21          | 3   | 1          | 5   | 18  |               |                                                                      |            |            |     |      |      |
|                                       |        | "           | 22  | 1          | 0   | 1   | 16            |                                                                      |            |            |     |      |      |
|                                       |        | Sums        | 61  | 27         | 40  | 235 |               |                                                                      |            |            |     |      |      |

Here again, we see a remarkable correspondence between the observed and the expected.

Thus, all of our expectations have been fulfilled and *it would be quite safe for us to conclude that our proposed gametic coupling between flowering time and flower colour is justified. And this conclusion will give strong support, in its turn, to our proposed two-factor hypothesis for the inheritance of the flowering time in peas.*

**SUMMARY.**

1. The fixity of the character of the flowering time in the original varieties of peas is established experimentally and the presence of two pure lines in the population of one variety is also established.
  2. The flowering time of the  $F_1$  is not intermediate between the parents, but inclines towards one of the parents ; in peas towards the late parent, and in rice towards the early parent.
  3. The variation range of  $F_2$  families covers the combined range of both parent varieties, but their variation type is not the ordinary one. Individuals, whose flowering time is just intermediate between those of the parents, are very few in number and sometimes absent (minimum frequency class), while in the ordinary variation type there should be the largest number of flowering individuals.
  4. By raising  $F_3$  and  $F_4$  families in peas, it is ascertained that the inheritance of the flowering time follows the Mendelian law.
  5. The inheritance of the flowering time in peas can be interpreted by proposing (1) the presence of two Mendelian factors which differ in their effects, and (2) gametic contamination caused by hybridization whose nature is not yet explainable.
  6. From variation types of  $F_1$ ,  $F_2$  and  $F_3$  families in peas, it is seen that the hereditary difference of the two pure lines is not quantitative but qualitative : i. e. the number of the factors is not different but the quality of the factors is different in both pure lines.
  7. Our material was not sufficient to propose a definite interpretation for the inheritance of shooting time in the rice varieties, but the hypothesis of three alleromorphic pairs does not positively conflict with the actual results, as far as our experiments go.
  8. The correlation between flower colour and flowering time in peas can be satisfactorily explained by assuming gametic coupling between colour factor and one of the two factors for flowering time.
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TABLE 9—Continued.

[illegible]





TABLE 10—Frequency distribution of number of days from sowing to flowering in  $F_2$  of crosses between *M. P.* and early line of *G. P.* (1912).

| No. | Parent Designation            | Parent Class | Class Centiles |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
|-----|-------------------------------|--------------|----------------|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|
|     |                               |              | 42             | 43 | 44 | 45 | 46 | 47 | 48 | 49 | 50 | 51 | 52 | 53 | 54 | 55 | 56 | 57 | 58 | 59 | 60 | 61 | 62 | 63 |
|     | <i>M. P.</i> — <i>G. P.</i>   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
|     | <i>M. P.</i> — <i>G. P.</i>   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 1   | <i>I</i> × <i>I</i> 4-29.39   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 2   | <i>I</i> × <i>I</i> 4-24.53*  |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 3   | <i>I</i> × <i>I</i> 4-26.54   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 4   | <i>I</i> × <i>I</i> 4-1.56    |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 5   | <i>I</i> × <i>I</i> 4-12.45   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 6   | <i>I</i> × <i>I</i> 4-19.46   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 7   | <i>I</i> × <i>I</i> 4-8.44    |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 8   | <i>I</i> × <i>I</i> 4-3.49    |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 9   | <i>I</i> × <i>I</i> 4-27.66   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 10  | <i>4</i> × <i>XVI</i> 2-27.52 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 11  | <i>4</i> × <i>XVI</i> 2-1.55  |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 12  | <i>I</i> × <i>I</i> 4-17.56   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 13  | <i>I</i> × <i>I</i> 4-28.56   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 14  | <i>I</i> × <i>I</i> 4-16.56   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 15  | <i>I</i> × <i>I</i> 4-6.58    |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 16  | <i>I</i> × <i>I</i> 4-5.59    |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 17  | <i>I</i> × <i>I</i> 4-7.44    |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 18  | <i>I</i> × <i>I</i> 4-4.56    |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 19  | <i>I</i> × <i>I</i> 4-22.43   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 20  | <i>4</i> × <i>XVI</i> 2-18.54 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 21  | <i>4</i> × <i>XVI</i> 2-28.52 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 22  | <i>4</i> × <i>XVI</i> 2-8.43  |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 23  | <i>4</i> × <i>XVI</i> 2-25.46 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 24  | <i>4</i> × <i>XVI</i> 2-7.47  |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 25  | <i>4</i> × <i>XVI</i> 2-6.48  |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 26  | <i>4</i> × <i>XVI</i> 2-11.48 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 27  | <i>4</i> × <i>XVI</i> 2-10.48 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 28  | <i>I</i> × <i>I</i> 4-11.53   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 29  | <i>I</i> × <i>I</i> 4-23.69   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 30  | <i>4</i> × <i>XVI</i> 2-26.55 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 31  | <i>I</i> × <i>I</i> 4-10.63   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 32  | <i>4</i> × <i>XVI</i> 2-24.53 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 33  | <i>I</i> × <i>I</i> 4-21.54   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 34  | <i>I</i> × <i>I</i> 4-20.56   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 35  | <i>4</i> × <i>XVI</i> 2-3.53  |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 36  | <i>4</i> × <i>XVI</i> 2-5.55  |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 37  | <i>4</i> × <i>XVI</i> 2-23.53 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 38  | <i>I</i> × <i>I</i> 4-2.53    |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 39  | <i>4</i> × <i>XVI</i> 2-20.51 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 40  | <i>4</i> × <i>XVI</i> 2-22.54 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 41  | <i>4</i> × <i>XVI</i> 2-21.51 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 42  | <i>4</i> × <i>XVI</i> 2-22.54 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 43  | <i>I</i> × <i>I</i> 4-9.55    |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 44  | <i>4</i> × <i>XVI</i> 2-2.51  |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 45  | <i>I</i> × <i>I</i> 4-14.56   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 46  | <i>4</i> × <i>XVI</i> 2-4.55  |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 47  | <i>4</i> × <i>XVI</i> 2-13.54 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 48  | <i>4</i> × <i>XVI</i> 2-12.54 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 49  | <i>I</i> × <i>I</i> 4-15.53   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 50  | <i>I</i> × <i>I</i> 4-18.56   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 51  | <i>4</i> × <i>XVI</i> 2-14.54 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 52  | <i>4</i> × <i>XVI</i> 2-16.56 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 53  | <i>4</i> × <i>XVI</i> 2-19.68 |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 54  | <i>I</i> × <i>I</i> 4-25.69   |              |                |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |

\* ( ) Late-sprouted.







TABLE 11—Continued.

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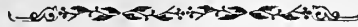
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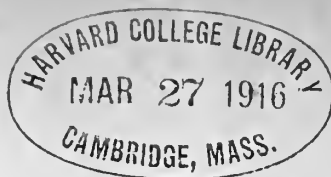
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# ÜBER DIE STICKSTOFFBESTANDTEILE VON PARALITHODES CAMTSCHATICA UND POLYPUS PUNCTATUS

VON

Eiji Takahashi, *Nōgakushi*

---

Die Krabben, welche neuerdings in beträchtlichen Mengen in Blechdosen verlötet aus Japan ausgeführt werden, gehören zur Art *Paralithodes camtschatica*, gemeiniglich in Japan Tarabagani genannt. Sie sind Bewohner der nördlichen Meere, weshalb bis jetzt keine Forschungen mit diesen Spezies im frischen Zustand ausgeführt worden sind.

Über die chemische Beschaffenheit des Krabbenfleisches liegen schon verschiedene Forschungen vor. So berichtet E. Röhrig<sup>1)</sup> vom Krabbenextrakt, dass 100 Teile desselben aus 23.98 Teilen Asche; 15.53 Teilen Kochsalz; 0.513 Teilen Stickstoff; 31.75 Teilen Rohprotein und 0.13 Teilen Phosphorsäure bestehen. D. Ackermann und F. Kutscher<sup>2)</sup> und später H. Barchall<sup>3)</sup> stellten darüber auch Untersuchungen an. Die ersteren stellten die Anwesenheit von Tyrosin, Leucin, Arginin, Lysin, Hypoxanthin, Betain, Neosin, Pyrimidinmethylechlorat, Phosphorsäure und zwei neulich gefundene Basen: Crangitin sowie Crängonin fest; der letztere bestimmte den Gesamtstickstoff auf 7.7; durch Phosphor-Wolframsäure fällbaren Stickstoff auf 2.7; Aminosäure-Stickstoff auf 0.90 und Ammoniak-Stickstoff auf 0.15 für 100 Teile des Extrakts. U. Suzuki im Verein mit Inouye und Bharatkar<sup>4)</sup> stellten ebenfalls Arginin, Histidin, Leucin, Tyrosin und Alanin fest

---

1) Zs. Unters. Nahrungsmittel., 12 (1906) p. 559.

2) Ibid. 13 (1907) I pp. 150-184, 610-614, II pp. 687-991.

3) Jahresb. f. Tierchemie 1910. p. 482.

4) Jour. Tokyo Chem. Soc. 31 (1909) p. 584.

und fanden überdies eine neue Base-Kanirin; die aus einer grossen Krabbenart gewonnen wurde, welche in der Provinz Echizen zuhause ist. Vor kurzem stellten auch Y. Okuda und H. Matsui<sup>1)</sup> über die Beschaffenheit von konserviertem Tarabaganiflesh Forschungen an und kamen zu dem Ergebnis, dass das Krabbenfleisch im Vergleich mit andern Fleischarten, mehr Nicht-eiweiss-Stickstoff und Schwefel enthält; und dass es durch Selbstverdauung, sowie bakterielle Wirkungen sich leicht basisch verändert und zu gleicher Zeit flüchtige Schwefelverbindungen auftreten lässt, wodurch während des Aufbewahrens Schwarzfärbung verursacht wird. H. Matsui sowie K. Nakazawa<sup>2)</sup> berichten auch über die Bestandteile von Hanasakigani (*Paralithodes brevipes* Brandt) und die chemischen Eigenschaften von Tarabagani in Beziehung zu ihrem Geschlecht. Die obigen Untersuchungen sind meistens mit Extrakt oder konservierten Präparaten durchgeführt worden; ich schätze mich deshalb glücklich, dass es möglich war, hier in Hokkaido die Versuche mit frischem Fleisch von Tarabagani auszuführen.

Die Stickstoffbestandteile der Mollusken sind schon durch U. Suzuki<sup>3)</sup> und seine Mitarbeiter für einige Arten d. h. Hamaguri, Ika und Austern untersucht und aus diesen die Basen Betain, Taurin, Leucin, Arginin etc. ausgefällt worden. In der vorliegenden Arbeit will ich über die Resultate meiner eigenen Forschungen, welche ich mit einer Tintenschneckenart (*Polypus punctatus* Gabb) angestellt habe, berichten.

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1) Jour. Scien. Agric. Soc., Tokyo 122 (1912) pp. 1-32.

2) Report of Fishery Inst. (Depart. agr. & commerce) Tokyo, Vol. 9, No. 5 (1914).

3) Jour. Tokyo Chem. Soc., 30 (1908) pp. 917-967.

# I. *Paralithodes camtschatica* (Tarabagani).

## Verteilung des Stickstoffes.

2200 gr des frischen Fleisches wurden mit Wasser bei 60° wiederholt extrahiert. Der gereinigte Auszug betrug 6.5 Liter und ergab bei der Analyse folgende Zusammensetzung:

| In 100 Teilen frischen Fleisches                     |       |
|------------------------------------------------------|-------|
| Gesamt-Stickstoff                                    | 2.710 |
| In Wasser löslicher Stickstoff                       | 1.26  |
| Davon entfielen auf                                  |       |
| Eiweiss-Stickstoff                                   | 0.308 |
| Ammoniak-Stickstoff                                  | 0.021 |
| Durch Phosphor-Wolframsäure fällbaren Stickstoff     | 0.342 |
| Stickstoff in anderer Form                           | 0.589 |
| In durch Phosphor-Wolframsäure fällbarem Stickstoff: |       |
| Durch Silbernitratfällbarer Stickstoff               | 0.013 |
| Durch Silbernitrat und Baryt fällbarer Stickstoff    | 0.109 |
| Sonstiger Stickstoff                                 | 0.218 |
| In Wasser löslicher Stickstoff 100                   |       |
| Eiweiss-Stickstoff                                   | 24.44 |
| Ammoniak-Stickstoff                                  | 1.67  |
| Durch Phosphor-Wolframsäure fällbarer Stickstoff     | 27.14 |
| Sonstiger Stickstoff                                 | 46.75 |
| Durch Phosphor-Wolframsäure fällbarer Stickstoff 100 |       |
| Durch Silbernitrat fällbarer Stickstoff              | 3.81  |
| Durch Silbernitrat und Baryt fällbarer Stickstoff    | 31.91 |
| Sonstiger Stickstoff                                 | 64.28 |

## Isolierung von organischen Basen.

6 Liter der in der oben angegebenen Weise dargestellten Auszüge wurden, nachdem sie mit Essigsäure schwach angesäuert waren, mit nicht zu viel 10% iger Tanninlösung versetzt und der entstandene Niederschlag nach 24 Stunden

abfiltriert. Das Filtrat wurde dann mit Bleiessig in schwachem Überschuss versetzt und der entstandene Niederschlag wieder abgesaugt. Das Filtrat vom Bleiessig-Niederschlag wurde mittels Schwefelsäure vom Blei befreit, mit derselben Säure zu ungefähr 5 % angesäuert und dann mit konzentrierter Lösung von Phosphor-Wolframsäure vollständig gefällt. Der dabei erhaltene Phosphor-Wolframsäureniederschlag wurde nach 24 stündigem Stehen mit Hilfe einer Nutsche von der Flüssigkeit getrennt und mit 5 % iger Schwefelsäure gewaschen. Der Niederschlag wurde nun mit überschüssigem Baryt verrieben. Das Gemisch wurde öfters umgerührt und 24 Stunden lang stehen gelassen und dann abgesaugt. Der Rückstand wurde nochmals in Wasser verteilt und mit Baryt verrieben. Diese Operation wurde also zweimal wiederholt. Die vereinigten Filtrate wurden durch Kohlensäure vom überschüssigen Baryt befreit und abfiltriert. Die dabei in Freiheit gesetzte Basenlösung wurde mit Salpetersäure neutralisiert und mit Silbernitrat in mässigem Überschuss versetzt, wobei ein gelbbrauner Niederschlag auftrat.

#### **A. Die durch Silbernitrat fällbaren Basen.**

Der Silbernitratniederschlag wurde zur Entfernung der Salpetersäure mit kaltem Wasser ausgewaschen, in einen Becher gespült und mit Überschuss von Ammoniak 24 Stunden lang digeriert, um die Silbernitratsalze der Basen in ihre Silbersalze überzuführen. Dann fügte ich dieser Ammoniaklösung etwas Silbernitrat hinzu, filtrierte nach dem Erkalten und wusch so lang mit kaltem Wasser aus, bis im Filtrat keine Trübung durch Chlorsilber mehr zu bemerken war. Nun wurden die reinen Silbersalze in Wasser suspendiert und mit warmer verdünnter Salzsäure versetzt. Die vom Chlorsilber abfiltrierte klare Flüssigkeit wurde stark eingeeengt, mit einem Überschuss von Ammoniak versetzt und 24 Stunden lang stehen gelassen, wobei ein schwach gelblicher Niederschlag ausgeschieden wurde, dessen Gesamtausbeute nach dem Eintrocknen jedoch weniger als 0,001 gr betrug. Aus diesem Grunde konnte ich weitere Versuche über die Beschaffenheit dieser Substanz leider nicht vornehmen; doch glaube ich, gestützt auf diesen Versuch, die Anwesen-



heit von Guanin annehmen zu dürfen.

Das ammoniakalische Filtrat wurde zur Vertreibung des Ammoniaks auf dem Wasserbade eingengt, mit Salzsäure angesäuert, wiederholt zur Trockne verdampft und zuletzt mit Alkohol verrieben. Der dabei ungelöst gebliebene Rückstand wurde in wenig Wasser gelöst. Die erhaltene Lösung wurde zur Isolierung sonstiger Purinbasen weiter behandelt, aber es wurden keine sicheren Ergebnisse erzielt.

### **B. Durch Silbernitrat und Baryt fällbare Basen.**

Der Mutterlauge vom Silbernitratniederschlag wurden noch Silbernitrat und Baryt im mässigen Überschuss zugesetzt, wobei ein dunkelbrauner Niederschlag in reichlicher Menge abgeschieden wurde. Dieser wurde nach dem Auswaschen mit sehr verdünntem Barytwasser in einer mit Schwefelsäure angesäuerten geringen Menge Wasser verteilt und durch Schwefelwasserstoff zersetzt. Das Filtrat von Schwefelsilber wurde nach Vertreibung des Schwefelwasserstoffs auf dem Wasserbade mittels Baryt von der Schwefelsäure und hernach mittels Kohlensäure vom überschüssigen Baryt befreit, worauf die Flüssigkeit mit soviel Schwefelsäure versetzt wurde, bis der Gehalt an Säure auf ungefähr 5 % stieg. Zum Schluss wurde die Mischung mit Phosphor-Wolframsäure wiederum gefällt. Der Phosphor-Wolframsäureniederschlag wurde in der oben beschriebenen Weise mit Baryt zersetzt und das überschüssige Baryt durch Kohlensäure entfernt. Im weitem wurde die vom Ammoniak befreite Lösung von isolierten Basen nach Sättigung mit Kohlensäure mit einer genügenden Menge von Quecksilberchlorid versetzt. Der entstandene Niederschlag wurde abgenutscht und mit Wasser gewaschen. Hierauf wurde derselbe in wenig Wasser verteilt und mit Schwefelwasserstoff zersetzt. Der dabei entstandene Schwefelquecksilberniederschlag wurde abfiltriert und mit Wasser gewaschen. Das Filtrat wurde im Vakuum eingedampft und im Vakuumexsikkator stehen gelassen, wobei sich prismatische Krystalle in geringer Menge ausschieden. Die Krystalle gaben mit Kupfersulphat und Natronlauge die Biuret-Reaktion und zeigten mit Diazobenzol-

sulfonsäure und Alkali tiefkirschrote Färbung. Die Ausbeute betrug 0.09 gr.

Für die Analyse wurde die Masse im Vakuum bei 100° getrocknet.

0.088 gr Substanz gaben      0.01592 gr N

N

Berechnet für  $C_6H_9N_3O_2 \cdot 2HCl$ :      18.42

Gefunden:      18.08

Gestützt auf die angegebenen Daten wurden die Krystalle als mit Histidinhydrochlorat identisch erkannt.

Das Filtrat vom oben erwähnten Quecksilberchloridniederschlag wurde durch Schwefelwasserstoff vom Quecksilber befreit, unter vermindertem Druck etwas eingedampft, um den überschüssigen Schwefelwasserstoff zu entfernen, und dann mit Silbernitrat von der Salzsäure befreit. Dem Filtrat wurde ein Überschuss von Silbernitrat und Baryt zugesetzt, wobei ein reichlicher Niederschlag entstand, welcher mit Hilfe der Saugpumpe abgesaugt wurde. Der Niederschlag wurde in schwefelsäurehaltigem Wasser verteilt, mit Schwefelwasserstoff zerlegt, darauf abfiltriert. Die Flüssigkeit wurde nach Neutralisierung mit Salpetersäure im Vakuum eingengt und mehrere Stunden lang stehen gelassen. Es schieden sich dabei feine Krystallnadeln ab, die von der Mutterlauge abfiltriert wurden. Nach dem Eintrocknen über Schwefelsäure im Vakuum, wurden sie gewogen und darnach analysiert. Die Ausbeute betrug 2 gr.

0.1 gr Substanz gab      0.8910 gr  $CO_2$       0.0482 gr  $H_2O$

C

H

Berechnet für  $C_6H_{14}N_4O_2 \cdot 2HNO_3$ :      23.98      4.70

Gefunden:      24.30      5.35

Aus dem obigen Analysenwert war es deutlich, dass die Krystalle Arginininitrat waren.

Das salpetersaure Salz wurde in wenig Wasser gelöst und mit einer gesättigten Lösung von Natriumpikrat versetzt, wobei sich gelbe Nadeln abschieden, die aus einer kleinen Menge Wasser nochmals krystallisiert wurden. Der Schmelzpunkt dieses Pikrats lag bei etwa 210°C (unkorr.). Für die

Analyse wurde es im Vakuum bei 100° getrocknet.

|                                                      |                    |             |
|------------------------------------------------------|--------------------|-------------|
| 0.3 gr Substanz gaben                                | 0.1684 Pikrinsäure |             |
|                                                      |                    | Pikrinsäure |
| Berechnet für $C_6H_{14}N_4O_2 \cdot C_6H_3N_3O_7$ : | 56.82              |             |
| Gefunden :                                           | 56.13              |             |

### C. Durch Silbernitrat und Baryt nicht fällbare Basen.

Nachdem die Mutterlauge des Silbernitrat- und Barytniederschlags durch Salzsäure vom Silber und durch Schwefelsäure vom Baryt befreit worden war, wurde sie mit dieser letztern Säure stark angesäuert und mit Phosphor-Wolframsäure versetzt, wobei ein weisser Niederschlag in reichlicher Menge auftrat. Der Niederschlag wurde mit 5 % iger Schwefelsäure gewaschen und mit Baryt zerlegt. Die erhaltene klare Basenlösung wurde im Vakuum zu Sirup verdunstet und im Vakuumexsikkator mehrere Stunden lang stehen gelassen, wobei sich grosse platte Krystalle ausschieden, welche nach Waschen mit absolutem Alkohol und Aether über Schwefelsäure getrocknet wurden. Die Ausbeute davon betrug 5.0 gr.

Die Krystalle waren sehr zerfliessbar, farblos und durchsichtig, sehr löslich in Wasser sowie heissem Alkohol, mit Pikrinsäure und Alkali zeigten sie keine Rotfärbung, ferner besitzen die Krystalle einen angenehmen süssen Geschmack.

Ein Teil der Krystalle wurde in heissem Aethylalkohol gelöst, mit Aether versetzt, und die Mischung eine kurze Weile stehen gelassen, wobei glänzende kleine tafelförmige Krystalle in reichlicher Menge ausgeschieden wurden, die auf einem Filtrierpapier gesammelt und mit Alkohol und Aether gewaschen wurden. Für die Analyse wurden sie aus Alkohol wiederum krystallisiert und im Vakuum bei 80° getrocknet.

|                               |                  |                  |       |
|-------------------------------|------------------|------------------|-------|
| 0.1306 gr Substanz gaben      | 0.2440 gr $CO_2$ | 0.1076 gr $H_2O$ |       |
| 0.3 gr Substanz gaben         | 0.03110 gr N     |                  |       |
|                               | C                | H                | N     |
| Berechnet für $C_5H_{11}NO_2$ | 51.28            | 9.40             | 11.97 |
| Gefunden :                    | 50.96            | 9.15             | 12.03 |

Um das Pikrat darzustellen, löste ich die erhaltenen Krystalle in einer kleinen Menge Wasser auf, fügte die gesättigte Pikrinsäurelösung hinzu, wobei sich nach einiger Zeit schöne prismenförmige Krystalle bildeten. Die Krystalle wurden gesammelt, mit Wasser gewaschen und durch nochmaliges Krystallisieren aus heissem Wasser gereinigt. Für die Analyse wurden sie im Vakuum bei 100° getrocknet.

0.3 gr Substanz gaben 0.04749 gr N

0.4121 gr Substanz gaben 0.2701 gr Pikrinsäure

|                                             | N     | Pikrinsäure |
|---------------------------------------------|-------|-------------|
| Berechnet für $C_5H_{11}NO_2C_6H_3N_3O_7$ : | 16.19 | 66.19       |
| Gefunden:                                   | 15.83 | 66.56       |

Im Kapillarrohr erhitzt, fangen die Krystalle gegen 170° an, zusammenzuschrumpfen, worauf sie gegen 182° (unkorr.) unter Zersetzung schmelzen.

Um auch das Chlorplatin doppelsalz zu gewinnen, wurde die Substanz in wenig mit Salzsäure schwach angesäuertem Wasser gelöst und im Vakuum über Schwefelsäure langsam eingeengt, wobei sich gelbe prismatische Krystalle ausschieden. Diese wurden mit Wasser gewaschen und aus mit Salzsäure angesäuertem Wasser wiederum in gelbe rhombische Tafeln umkrystallisiert, die bei Erhitzung im Kapillarrohr bei 246–247° (unkorr.) sich zersetzten. Für die Analyse wurde die Substanz im Vakuum bei 100° getrocknet.

0.1247 gr Substanz gaben 0.0375 gr Pt.

|                                           | Pt.   |
|-------------------------------------------|-------|
| Berechnet für $(C_5H_7NO_2HCl)_2PtCl_4$ : | 30.29 |
| Gefunden:                                 | 30.09 |

Gestützt auf alle diese Eigenschaften und die Analysenwerte wurden die Krystalle als mit Betain vollständig identisch erkannt. Die von U. Suzuki bei grössern Krabben gefundene Base Kanirin konnte ich aus dieser Spezies nicht gewinnen.

**D. Basen im Filtrat vom Phosphor-Wolframsäureniederschlag**

Aus dem Filtrat vom Phosphor-Wolframsäureniederschlag wurde mit Baryt die Schwefelsäure, sowie die Phosphor-Wolframsäure beseitigt; sodann wurde mit Schwefelsäure die Lösung sorgfältig vom überschüssigen Baryt befreit und unter sehr geringem Druck zu geringem Volumen verdunstet. Aus dieser Lösung schieden sich die Krystallmassen ab. Dieselben wurden nach 48 Stunden scharf abgesaugt und mit Alkohol und Aether gewaschen. Sie stellten, nach ihrer Gestalt zu schliessen, ein Gemenge von Tyrosin und Leucin dar, das deshalb durch mit Alkohol vermischtem Eisessig in seine Teil getrennt wurde. Die beiden Körper wurden durch ihre Reaktionen und die Analyse identifiziert.

|                                                                |                             |                            |
|----------------------------------------------------------------|-----------------------------|----------------------------|
| 0.12 gr Tyrosin gaben                                          | 0.261 gr CO <sub>2</sub> .  | 0.0667 gr H <sub>2</sub> O |
|                                                                | C                           | H                          |
| Berechnet für C <sub>9</sub> H <sub>11</sub> NO <sub>3</sub> : | 59.66                       | 6.07                       |
| Gefunden :                                                     | 59.30                       | 6.16                       |
| 0.1008 gr Leucin gaben                                         | 0.2017 gr CO <sub>2</sub> , | 0.088 gr H <sub>2</sub> O  |
|                                                                | C                           | H                          |
| Berechnet für C <sub>6</sub> H <sub>13</sub> NO <sub>2</sub>   | 54.96                       | 9.92                       |
| Gefunden :                                                     | 54.57                       | 9.70                       |

Die ganze Ausbeute an Tyrosin betrug 0.28 gr, die an Leucin etwa 1 gr.

**Zusammenfassung der Ergebnisse.**

Aus 2 kg frischem Krabbenfleische wurden an Basen folgende Mengen gewonnen :

|                        |           |
|------------------------|-----------|
| Histidin (Chlorhydrat) | 0.082     |
| Arginin (Nitrat)       | 1.81      |
| Betain                 | 4.00      |
| Tyrosin                | 0.25      |
| Leucin                 | 0.92      |
| Purin Basen            | Vorhanden |

Wir erschen daran, dass die erhaltenen Resultate einigermaßen mit denen früherer Versuche nicht übereinstimmen. Die Bestandteile weisen, vielleicht den Arten entsprechend, bedeutende Abweichungen auf.

## II. *Polypus punctatus* Gabb (Tintenschnecke).

2 Kilo frisches, von Haut und Saugnäpfen befreites Fleisch von Tintenschnecken wurde erst möglichst stark zerkleinert und dreimal mit Wasser (50–60°) extrahiert. Die Auszüge massen im ganzen 4.5 Liter. Die quantitative Bestimmung des Extraktes ergab folgendes Resultat:

|                    | In 100 cc Extrakt | In 100 Teilen<br>Trockensubstanz des Extraktes |
|--------------------|-------------------|------------------------------------------------|
| Trockensubstanz    | 3.530             | 100.00                                         |
| Gesamtstickstoff   | 0.350             | 9.91                                           |
| Eiweisstickstoff   | 0.063             | 1.78                                           |
| Basenstickstoff    | 0.140             | 1.97                                           |
| Ammoniakstickstoff | Spur              | —                                              |

|                       | In 100 Teilen frischem Fleisch |
|-----------------------|--------------------------------|
| Lösliche Substanz     | 7.942                          |
| Löslicher Stickstoff  | 0.787                          |
| Löslicher Eiwei-stoff | 0.142                          |
| Basenstickstoff       | 0.315                          |

### A. Isolierung von Taurin.

1.5 Liter der wässerigen Auszüge wurden mit 10% iger Tanninlösung versetzt und der entstandene Niederschlag abfiltriert. Das Filtrat wurde nach dem Entfernen des Tannins durch Bleiessiglösung und des überschüssigen Bleis durch Schwefelsäure im Vakuum stark eingeeengt und dann einige Wochen lang im Exsikkator sich selbst überlassen, wobei sich grosse glänzende monokline Prismen ausschieden, die ungefähr 2.5 gr betrugen. Die resultierenden Krystalle waren in Wasser leicht, in absolutem Alkohol gar nicht löslich. Der qualitative Nachweis ergab das Vorhandensein von

Schwefel. Für die Analyse wurden die Krystalle aus heissem Wasser umkrystallisiert und bei  $100^{\circ}$  getrocknet.

|                                                                |                             |       |
|----------------------------------------------------------------|-----------------------------|-------|
| 0.3115 gr Substanz gaben                                       | 0.03548 gr N                |       |
| 0.1806 gr Substanz gaben                                       | 0.3487 gr BaSO <sub>4</sub> |       |
|                                                                | N                           | S     |
| Berechnet für C <sub>2</sub> H <sub>7</sub> NSO <sub>3</sub> : | 11.20                       | 25.60 |
| Gefunden :                                                     | 11.39                       | 25.41 |

Die Resultate stimmen mit den Eigenschaften von Taurin ziemlich gut überein.

### B. Isolierung von organischen Basen.

Ferner wurden 2.5 Liter des Originalauszuges nach der Behandlung mit Tannin und Bleiessiglösung in oben beschriebener Weise mit Schwefelsäure versetzt, bis der Gehalt davon 5 % betrug und dann durch Phosphor-Wolframsäure ausgefällt. Der erhaltene Niederschlag wurde in bekannter Weise mit Baryt zerlegt und das überschüssige Baryt durch Kohlensäure entfernt. Die alkalische Lösung, welche freie Basen enthielt, wurde hierauf mit Kohlensäure gesättigt und mit Quecksilberchlorid gefällt.

a) Der Quecksilberchloridniederschlag wurde gut gewaschen, in wenig Wasser verteilt und mit Schwefelwasserstoff zerlegt. Das Filtrat wurde darauf im Vakuum zu einer kleinen Menge eingeeengt. Dem so gewonnenen Sirup wurde eine genügende Menge von absolutem Methylalkohol zugefügt und trockenes Salzsäuregas zunächst bis zur Sättigung eingeleitet, wodurch die Base in Methylesterhydrochlorat umgewandelt wurde. Durch Verdampfen des Alkohols schieden sich farblose prismatische Krystalle aus, welche aus heissem Alkohol unter Zusatz von Aether umkrystallisiert, im Vakuum bei  $100^{\circ}$  getrocknet und hierauf analysiert wurden. Gesamt-Gewicht der Krystalle betrug 0.6 gr.

|                          |                             |                            |
|--------------------------|-----------------------------|----------------------------|
| 0.2476 gr Substanz gaben | 0.3176 gr CO <sub>2</sub> , | 0.1275 gr H <sub>2</sub> O |
| 0.2000 g. Substanz gaben | 0.2362 gr AgCl              |                            |

|                                            | C     | H    | Cl    |
|--------------------------------------------|-------|------|-------|
| Berechnet für $C_6H_8N_3O_2 (CH_3)_2HCl$ : | 34.67 | 5.45 | 29.30 |
| Gefunden:                                  | 34.99 | 5.72 | 29.08 |

Aus dem Analysenwert ergab es sich, dass die erhaltenen Krystalle aus Histidinmethylesterhydrochlorat bestanden.

Die Krystalle waren sowohl in Wasser, als auch in Methylalkohol leicht-, dagegen in Aether unlöslich.

Sie zeigten als wässrige Lösung zusammen mit alkalischer Diazobenzolsulfonsäure rote Färbung, mit Millon'scher Reagenz trat die Ausscheidung eines weissen Niederschlags auf. Der Schmelzpunkt lag bei  $198^\circ$  (unkorr.). Diese Eigenschaften stehen mit denen von Histidinmethylesterhydrochlorat ganz in Übereinstimmung.

b) Das Filtrat vom Quecksilberniederschlag des Histidins wurde mittels Schwefelwasserstoff vom Quecksilber und, nachdem der überschüssige Schwefelwasserstoff im Vakuum entfernt war, mit Silbernitrat auch von der Salzsäure befreit, alsdann wurde die Lösung mit einem Überschuss von Silbernitrat und Baryt gefällt. Der Niederschlag wurde durch Schwefelwasserstoff zerlegt und abfiltriert. Die Flüssigkeit wurde im Vakuum bedeutend eingedampft und mit gesättigter Lösung von Pikrinsäure versetzt, wodurch eine ziemlich beträchtliche Ausscheidung von feinen seidenglänzenden Krystallen erfolgte. Die Krystalle wurden dann über Schwefelsäure getrocknet und gewogen. Die Ausbeute betrug 2.69 gr. Für die Analyse wurden die Krystalle aus wenig Wasser umkrystallisiert und im Vakuum bei  $100^\circ$  getrocknet.

|                          |                       |
|--------------------------|-----------------------|
| 0.2561 gr Substanz gaben | 0.05457 gr N          |
| 0.2279 gr Substanz gaben | 0.1137 gr Pikrinsäure |

|                                                    | N     | Pikrinsäure |
|----------------------------------------------------|-------|-------------|
| Berechnet für $C_9H_{11}N_4O_3 \cdot C_6H_3N_3O_7$ | 21.54 | 50.33       |
| Gefunden:                                          | 21.23 | 49.89       |

Im Kapillarrohr erhitzt, färbten sie sich gegen  $200^\circ C$  braun und schmolzen gegen  $218^\circ$  (unkorr.) unter Zersetzung.

Aus dem beobachteten Schmelzpunkt und dem Analysenwert geht mit



Bestimmtheit hervor, dass das isolierte Pikrat nichts anderes ist als das Carnosin-pikrat.

Um das Nitrat zu gewinnen, wurde ein Teil des Pikrats in einer geringen Menge Wasser gelöst und mit etwas überschüssiger Salzsäure versetzt, wobei die ausgeschiedene Pikrinsäure entfernt wurde. Nachdem noch die gebliebene Pikrinsäure durch Aether entfernt worden war, wurde die Mutterlauge durch Phosphor-Wolframsäure gefällt. Der Niederschlag wurde durch Baryt zersetzt und die Lösung daraufhin mit Salpetersäure genau neutralisiert, im Vakuum eingengt und im Exsikkator stehen gelassen, wobei sich farblose prismatische Krystalle ausschieden. Diese wurden mit Alkohol gewaschen und aus Wasser umkrystallisiert. Für die Analyse wurden sie im Vakuum bei 100° getrocknet.

0.2678 gr Substanz gaben      0.3440 gr Nitronnitrat

|                                                                                                 |                  |
|-------------------------------------------------------------------------------------------------|------------------|
|                                                                                                 | HNO <sub>3</sub> |
| Berechnet für B <sub>9</sub> H <sub>14</sub> N <sub>4</sub> O <sub>3</sub> · HNO <sub>3</sub> : | 21.80            |
| Gefunden:                                                                                       | 21.58            |

Der Schmelzpunkt dieser Krystalle lag bei 210° (unkorr.).

c) Das Filtrat vom Silbernitrat- und Barytniederschlag wurde nach dem Entfernen des Silbers durch Salzsäure und des Überschusses von Baryt durch Schwefelsäure etwas eingengt und wieder mit Phosphor-Wolframsäure ausgefällt. Der Niederschlag wurde in bekannter Weise mit Baryt zerlegt. Das Filtrat vom Bariumsalz wurde nach dem Entfernen des Überschusses von Baryt durch Kohlensäure zu Sirup eingedampft und mehrere Tage im Exsikkator stehen gelassen, wodurch farblose tafelförmige Krystalle entstanden, die 3.65 gr wogen. Sie besitzen sehr süßen Geschmack, sind in Wasser sowie Alkohol sehr leicht, in Aether jedoch nicht löslich. Die Krystalle wurden sodann in heissem Alkohol aufgelöst und unter Zusatz von Aether umkrystallisiert. Für die Analyse wurde die Substanz im Vakuum bei ca 80° getrocknet.

0.2500 gr Substanz gaben      0.4673 gr CO<sub>2</sub>,      0.2083 gr H<sub>2</sub>O

|                                 | C     | H    |
|---------------------------------|-------|------|
| Berechnet für $C_5H_{11}NO_2$ : | 51.28 | 9.40 |
| Gefunden :                      | 50.98 | 9.26 |

Alle diese Eigenschaften und der Analysenwert stimmen mit denen des Betains überein.

Zur Bereitung der Pikrats wurde ein Teil der Krystalle in geringer Menge Wasser gelöst und mit gesättigter Pikrinsäurelösung versetzt, dabei schieden sich schöne Krystalle ab.

Dieselben wurden aus heissem Wasser umkrystallisiert, im Vakuum bei  $100^\circ$  getrocknet und darauf analysiert.

|                                                    |                    |
|----------------------------------------------------|--------------------|
| 0.3008 gr Substanz gaben                           | 0.1998 Pikrinsäure |
|                                                    | Pikrinsäure        |
| Berechnet für $C_5H_{11}NO_2 \cdot C_6H_3N_3O_7$ : | 66.19              |
| Gefunden                                           | 66.42              |

Das Pikrat schmolz bei Erhitzung im Schmelzröhrchen bei genau  $182^\circ$  (unkorr.).

### Zusammenfassung der Resultate.

Aus 2 kg frischem Fleisch wurden folgende Mengen Stickstoffverbindungen gewonnen :

|                                    |         |
|------------------------------------|---------|
| Taurin                             | 7.46 gr |
| Carnosin (Pikrat)                  | 4.79 gr |
| Betain                             | 6.57 gr |
| Histidin (Methylesterhydrochlorat) | 1.08 gr |



# ÜBER DAS VORKOMMEN VON BETAIN IN EINIGEN MEERESTIEREN

VON

Eiji Takahashi, *Nōgakushi*

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Das Betain ist bis jetzt häufig in Pflanzen gefunden worden, dagegen besitzen wir nur einige Berichte über die Verteilung dieser Substanz im Tierreich. Brieger<sup>1)</sup> fand sie zuerst im wässrigen Auszug der Miesmuschel (*Mytilus edulis*), D. Ackermann und F. Kutscher<sup>2)</sup> im Krabben-Extrakt, nachher U. Suzuki<sup>3)</sup> und seine Mitarbeiter in Austern, Tintenfischen (*Onustrephes* sp., im Aal (*Anguilla fluviatilis*) und der Muschel Hamaguri (*Cytherea meretrix* L.); ferner konnte der Autor<sup>4)</sup> die Substanz auch aus einer Krabbenart (*Paralithodes camtschatica*) und einer Tintenschnecke (*Polypus punctatus*) isolieren.

Das Betain besitzt einen stark süßen Geschmack, und da diese Substanz unzweifelhaft im freien Zustand vorhanden ist, erklärt sich auch der bei mehreren Arten vorhandene süsse Geschmack dieser Meerestiere. In meinen Untersuchungen versuchte ich den Nachweis des Vorkommens von Betain für folgende süßschmeckende drei Arten: Hummer und zwei Muschelarten, wobei eine nicht unbeträchtliche Menge der Substanz hergestellt wurde.

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1) Ptomain III p. 76.

2) Zs. Unters. Nahrungsmittel., **13** (1907). p. 610.

3) Jour. Tokyo Chem. Soc., **30** (1909) pp. 917-967.

4) Siehe vorgängige Beschreibung!

### I. Hokki (*Macra sachalinensis* Schrenck)

1200 gr des vollkommen frischen Fleisches der Muschel wurden in kleine Stücke zerschnitten, mit Wasser bei 50–60° extrahiert und abgepresst. Der Rückstand wurde in derselben Weise dreimal behandelt und etwa 3 Liter als Gesamtauszug erhalten. Die Flüssigkeit wurde sodann mit Essigsäure schwach angesäuert, mit konzentrierter Tanninlösung versetzt, worauf der dabei entstandene Niederschlag abfiltriert wurde. Das erhaltene Filtrat wurde mit Bleiessig vom überschüssigen Tannin, sowie anderen Eiweissstoffen befreit, ferner wurde durch Schwefelsäure das Blei ausgeschieden. Hierauf wurde die Flüssigkeit mit derselben Säure zu 5 % angesäuert und mit Phosphor-Wolframsäure gefällt. Der Niederschlag wurde mit Baryt in der allgemein üblichen Weise zersetzt, mit Schwefelsäure das Baryt beseitigt, sodass dann durch Filtrieren die klare Basenlösung gewonnen werden konnte. Die Lösung wurde dann weiterhin von dem durch Behandlung mit Silbernitratlösung ausgeschiedenen Niederschlag befreit, dazu überschüssige Silbernitrat- und Barytlösung addiert, wornach das entstandene Fällungsprodukt abermals abfiltriert wurde. Das Filtrat wurde durch Salzsäure und Schwefelsäure von Silber und Baryt befreit, mit Schwefelsäure stark angesäuert und wiederum mit Phosphor-Wolframsäure vollständig gefällt. Der gewonnene Niederschlag wurde mit Baryt zersetzt und das überschüssige Baryt durch Kohlensäure entfernt. Die in Freiheit gesetzte Basenlösung wurde im Vakuum sorgfältig bei niedriger Temperatur verdunstet, wobei zerfließbare tafelförmige Krystalle entstanden. Dieselben wurden mit absolutem Alkohol und Aether gewaschen und über Schwefelsäure getrocknet. Die Gesamtausbeute betrug etwa 5 gr.

Die Krystalle waren farblos und durchsichtig, leicht löslich in Wasser und Methylalkohol, etwas schwerer im Äthylalkohol und ganz unlöslich in Äther. Sie gaben weder die Diazo- noch die Biuret-Reaktion und zeigten weiter bei Erwärmen mit Kupferoxyhydrat keine blaue Färbung im Unterschied zu Aminosäuren. Im Vergleich mit dem aus Krabbenfleisch hergestellten Betain stimmten ihre physikalischen und chemischen Eigenschaften

ganz überein.

Für die Analyse wurde die Substanz aus heissem absolutem Aethylalkohol durch Aether krystallisiert und im Vakuum bei 80° getrocknet.

0.1455 gr Substanz gaben 0.2742 gr CO<sub>2</sub>, 0.1209 gr H<sub>2</sub>O

0.3 gr Substanz gaben 0.0348 gr N

|                                                                | C     | H    | N     |
|----------------------------------------------------------------|-------|------|-------|
| Berechnet für C <sub>5</sub> H <sub>11</sub> NO <sub>2</sub> : | 51.28 | 9.40 | 11.97 |
| Gefunden:                                                      | 51.36 | 9.23 | 11.63 |

Zur Bereitung des Pikrats wurde eine kleine Menge der umkrystallisierten Substanz in geringer Menge Wasser gelöst, mit gesättigter Pikrinsäurelösung versetzt, wobei sich schöne gelbe Krystalle abschieden. Dieselben wurden aus heissem Wasser umkrystallisiert, im Vakuum bei 100° getrocknet und darauf analysiert.

0.3501 gr Substanz gaben 0.2301 gr Pikrinsäure

|                                                                                                                            | Pikrinsäure |
|----------------------------------------------------------------------------------------------------------------------------|-------------|
| Berechnet für C <sub>5</sub> H <sub>11</sub> NO <sub>2</sub> C <sub>6</sub> H <sub>3</sub> N <sub>3</sub> O <sub>7</sub> : | 66.19       |
| Gefunden:                                                                                                                  | 65.72       |

Die umkrystallisierten Krystalle schieden sich als kleine Stäbchen aus; im Schmelzröhrchen erhitzt, schmolzen sie bei 182° (unkorr.).

Chlorplatin doppelsalz wurde ebenso gewonnen. Zu diesem Zweck wurden die Krystalle in wenig Wasser gelöst und mit verdünnter Salzsäure neutralisiert. Darauf fügte ich Chlorplatinlösung hinzu und engte die Lösung im Vakuumexsikkator ein. Die abgeschiedenen kleinen Krystalle wurden durch nochmaliges Krystallisieren aus wenig Wasser gereinigt. Sie zeigten die Form gelblicher rhombischer Tafeln. Im Kapillarrohr erhitzt, zersetzten sie sich unter Schwarzfärbung bei 246°–247° (unkorr.). Für die Analyse wurde die Masse im Vakuum bei 100° getrocknet.

0.1191 gr Substanz gaben 0.0365 gr Pt.

|                                                                                                     |       |
|-----------------------------------------------------------------------------------------------------|-------|
| Berechnet für (C <sub>5</sub> H <sub>11</sub> NO <sub>2</sub> HCl) <sub>2</sub> PtCl <sub>4</sub> : | 30.29 |
| Gefunden:                                                                                           | 30.67 |

Aus diesen Daten ergab sich deutlich, dass die isolierten Krystalle nichts anderes als Betain waren.

## II. Hummer (*Pandalus* sp.)

Über die basischen Bestandteile vom Hummer wurden vor kurzem von U. Suzuki und Y. Irie<sup>1)</sup> mit Iseyebi (*Panulirus* sp.) Untersuchungen angestellt und bewiesen, dass in demselben Arginin, Lysin, Histidin, Leucin, Tyrosin, Alanin und Prolin enthalten sind. Da es aber an einer Bemerkung über Betain mangelt, habe ich hier mit *Pandalus* sp. Untersuchungen vorgenommen.

1200 gr des Fleisches wurden zuerst möglichst fein zerkleinert und im Wassertrockenschrank bei etwa 80° getrocknet. Nachdem das Fleisch ziemlich getrocknet war, wurde es sorgfältig pulverisiert und durch ein feinmaschiges Sieb getrieben. Hierauf mit Aether von Fett und Farbstoffen befreit, mit rektifiziertem Alkohol 30 Minuten lang im Wasserbade gekocht und abgepresst. Diese Operation wurde also zweimal wiederholt. Das gereinigte Filtrat wurde unter vermindertem Druck zu einer kleinen Menge verdunstet, um den Alkohol zu vertreiben. Der Rückstand wurde mit Wasser aufgenommen. Zu dieser wässrigen Lösung fügte ich Tanninlösung, worauf der entstandene Niederschlag abgesaugt wurde. Das Filtrat wurde dann mit Bleiessig vom überschüssigen Tannin befreit und aus dem Filtrat mittels Schwefelsäure das Blei entfernt. Die Mutterlauge wurde nach Ansäuerung mit derselben Säure mit Phosphor-Wolframsäure gefällt und der Niederschlag mit Baryt zersetzt. Die erhaltene Basenlösung wurde zuerst mit Silbernitratlösung versetzt, der entstandene Niederschlag abfiltriert, und nachher der durch die Silbernitrat- und Barytlösung ausgeschiedene Niederschlag auch entfernt. Das Filtrat wurde mit Salzsäure und Schwefelsäure vom Silber und Baryt befreit, wiederum mit Phosphor-Wolframsäure gefällt und der Niederschlag mit Baryt zersetzt. Die Lösung wurde im Vakuum verdunstet und im Exsikkator stehen gelassen, wobei sich rhombische tafelförmige Krystalle abschieden. Dieselben wurden mit Alkohol und Aether gewaschen und über Schwefelsäure getrocknet. Die Ausbeute betrug 2.0 gr.

1) Jour. Tokyo Chem. Soc., 30 (1909) pp. 930-942.

Die Krystalle waren auch farblos und zerfliessbar, unlöslich in Aether, leicht löslich in Methylalkohol und warmen Aethylalkohol, ergaben weder Diazo- noch Biuretreaktion und zeigten sehr süssen Geschmack.

Für die Analyse wurden die Krystalle aus heissem Aethylalkohol umkrystallisiert und im Vakuum bei 80° getrocknet.

|                                                                |                           |                             |
|----------------------------------------------------------------|---------------------------|-----------------------------|
| 0.1 gr Substanz gaben                                          | 0.1888 gr CO <sub>2</sub> | 0.0871 gr. H <sub>2</sub> O |
|                                                                | C                         | H                           |
| Berechnet für C <sub>5</sub> H <sub>11</sub> NO <sub>2</sub> : | 51.28                     | 9.40                        |
| Gefunden :                                                     | 51.50                     | 9.68                        |

Das Pikrat wurde nach der oben geschilderten Weise aus dem umkrystallisierten Präparat dargestellt. Sein Schmelzpunkt lag bei 181–182° (unkorr.). Für die Analyse wurde es im Vakuum bei 100° getrocknet.

|                                                                                                                            |              |       |
|----------------------------------------------------------------------------------------------------------------------------|--------------|-------|
| 0.2582 gr Substanz gaben                                                                                                   | 0.03076 gr N |       |
|                                                                                                                            |              | N     |
| Berechnet für C <sub>5</sub> H <sub>11</sub> NO <sub>2</sub> C <sub>6</sub> H <sub>3</sub> N <sub>3</sub> O <sub>7</sub> : |              | 16.19 |
| Gefunden :                                                                                                                 |              | 15.79 |

Zur Umwandlung in das Nitrat wurden 0.5 gr der umkrystallisierten Substanz mit 5 cc normaler Salpetersäure versetzt und langsam, bis die Mischung ungefähr getrocknet war, verdampft. Die dabei entstandenen Krystalle wurden mit absolutem Alkohol verrührt, auf dem Filter gesammelt und mit Alkohol und Aether gewaschen. Für die Analyse wurde das Nitrat abermals aus heissem absolutem Alkohol krystallisiert und im Vakuum bei 100° getrocknet.

|                                                                                                 |                     |                  |
|-------------------------------------------------------------------------------------------------|---------------------|------------------|
| 0.2120 gr Substanz gaben                                                                        | 0.2665 Nitronnitrat |                  |
|                                                                                                 |                     | HNO <sub>3</sub> |
| Berechnet für (C <sub>5</sub> H <sub>11</sub> NO <sub>2</sub> ) <sub>2</sub> HNO <sub>3</sub> : |                     | 21.21            |
| Gefunden :                                                                                      |                     | 21.13            |

Dieses Sal z löst sich leicht in Wasser, dagegen etwas schwerer in Aethylalkohol, in Aether jedoch gar nicht auf. Aus heissem Aethylalkohol wird es durch Aether in farblose Prismen umkrystallisiert. Aus ihrer Beschaffenheit sowie dem Analysenwert zu schliessen, müssen die isolierten Krystalle mit Betain vollständig identisch sein.

### III. Hotate (*Pecten yessoensis* Jay)

1500 gr des fein zerschnittenen frischen Fleisches von Hotate wurden mit Wasser von 60° extrahiert und die gereinigten Auszüge mit Tannin, Bleiessig und Phosphor-Wolframsäure behandelt, so wie es mit Hokki der Fall war. Die erhaltene Lösung wurde dann von den durch Silbernitrat, sowie den durch Silbernitrat und Baryt fällbaren Basen befreit. Beim Abdampfen der Mutterlauge unter geringem Druck wurde eine Krystallmasse erhalten, welche sogleich in Aethylalkohol gelöst und durch Aether umkrystallisiert wurde. Durch nochmaliges Krystallisieren aus Alkohol und Entfärbung mit Tierkohle konnte ich die reine Substanz darstellen, deren Ausbeute etwa 0.7 gr betrug.

Die Krystalle besaßen mit dem aus Hummer oder Hokki isolierten Betain grosse Ähnlichkeit, waren farblos und zerfließbar, von sehr süßem Geschmack. Für die Analyse wurden sie im Vakuum bei 80° getrocknet.

0.1 gr Substanz gaben      0.1865 gr CO<sub>2</sub>,      0.0893 gr H<sub>2</sub>O

0.2 gr Substanz gaben      0.02367 gr N.

|                                                                | C     | H    | N     |
|----------------------------------------------------------------|-------|------|-------|
| Berechnet für C <sub>5</sub> H <sub>11</sub> NO <sub>2</sub> : | 51.28 | 9.40 | 11.97 |
| Gefunden:                                                      | 50.87 | 9.92 | 12.34 |

Das Pikrat wurde auch dargestellt, es schmolz gegen 182° (unkorr.).

Für die Analyse wurde es im Vakuum bei 100° getrocknet.

0.25 gr Substanz gaben      0.1670 gr Pikrinsäure

|                                                                                                                            | Pikrinsäure |
|----------------------------------------------------------------------------------------------------------------------------|-------------|
| Berechnet für C <sub>5</sub> H <sub>11</sub> NO <sub>2</sub> C <sub>6</sub> H <sub>3</sub> N <sub>3</sub> O <sub>7</sub> : | 66.19       |
| Gefunden:                                                                                                                  | 66.81       |

Der Analysenwert stimmt auch mit dem vom Betainpikrat überein.

Wie man oben ersehen kann, ist die Menge des aus Hotate dargestellten Betains geringer als 1 gr und entspricht gar nicht den Erwartungen. Ich erklärte dies mit der Tatsache, dass der süße Geschmack des Hotatefleisches nur einigen kleinen Teilen der Substanz eigen ist.



**Zusammenfassung der Resultate.**

Aus je 1 kg frischem Fleisch wurden folgende Mengen Betain gewonnen:

|                                                            |        |
|------------------------------------------------------------|--------|
| Hummer ( <i>Pudulus sp.</i> )                              | 2.0 gr |
| Hokki ( <i>Macra sachalinensis</i> Schrenk)                | 5 0 gr |
| Hotate ( <i>Pecten yessoensis</i> Jay)                     | 0.6 gr |
| Krabben ( <i>Paralithodes camtschitica</i> <sup>1)</sup> ) | 2.4 gr |
| Tintenschnecke ( <i>Polypus punctatus</i> )                | 3.7 gr |

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Zum Schluss möchte ich Herrn Prof. Dr. K. Oshima für seinen mir freundlichst gewährten Rat meinen herzlichsten Dank aussprechen.

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1) Siehe vorgängige Beschreibung!





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